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A CYTOLOGICAL STUDY OF PUCCINIA GLUMARUM ON BROMUS MARGINATUS AND TRITICUM VULGARE[1]

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INTRODUCTION

Stripe rust, *Puccinia glumarum* (Schm.) Eriks. and Henn., was discovered in America in 1915, and a series of investigations of the rust have been carried on here since that date. Humphrey, Hungerford, and Johnson (*23*)[3] published in 1924 an account of its discovery in America, its geographic range and host range, its relations to climate, and its economic importance. Hungerford (*24*) recorded data on overwintering and oversummering, viability of spores, and experiments proving that the disease "is not transmitted from one wheat crop to the next by means of infected seed wheat." Hungerford and Owens (*25*) in 1923 carried on extensive inoculations of *P. glumarum* on grasses and grains, particularly wheat, to determine the host range and physiologic forms of the rust and also to determine degrees of susceptibility and resistance in the hosts. The present paper adds to these investigations of stripe rust in America a cytological study of the uredinial and telial stages.

In "Die Getreideroste" in 1896, Eriksson and Henning (*20*) recounted their experiments which led to the division of *Puccinia rubigo-vera* (DC.) Wint. into several species, one of which was *P. glumarum*. This account included a description of *P. glumarum* with details of uredinia and telia on leaves, stems, heads, and seed of wheat. They found several distinct physiologic forms of the rust on different grains and grasses.

The mode of overwintering of the rust was obscure. The teliospores could germinate either in the fall or in the following spring. No aecial host was discovered, and the authors doubted the existence of an alternate host. Attempts to infect wheat with sporidia failed, but the authors did not consider the possibility excluded. Stripe rust is very resistant to cold, but its winter survival in Sweden by either urediniospores or uredinio-mycelium on young wheat plants was too rare to explain the sudden and widespread outbreaks of rust in the spring. Moreover, they found the germination of uredinio-

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[3] Reference is made by number (italic) to "Literature cited," p. 511.

spores to be poor and capricious ("launenhaft"), and believed that the spread of the rust from any one infection center was very limited. This led them to suggest the possibility of dormant mycelium in the seed as a means of overwintering and an explanation of the outbreaks of rust in the spring.

In pursuance of this idea, Eriksson (*14*, *15*) grew grain from rusted seed in small glass cases to prevent infection from without. Notwithstanding this, stripe rust appeared on the inclosed plants. No mycelium was found in very young seedlings, however, and Eriksson concluded that the "Krankheitsstoff" must be hidden and latent in the host cells in the form of an intimate symbiotic mixture of fungous and host protoplasm which he called "Mycoplasma." Only when outer conditions—soil and weather—were favorable did the fungous plasm separate itself from the host protoplasm, leave the host cells, and form ordinary intercellular mycelium.

Drawings representing the corpuscles of fungous plasm ready to leave the host cells were published in 1902 (*16*). These were followed by a series of illustrated cytological studies (*17*) giving details of the "dickem Plasma" in host cells near the borders of infections; of the swelling and dissolution of the host nucleus due to the fungus; the formation of "Plasmanucleoli" of fungous matter; the emptying of these nucleoli into intercellular spaces through fine tubes, giving rise to irregular masses of fungous plasm without septa and without nuclei; and finally of the formation of ordinary hyphae from these formless intercellular masses. The hyphae figured are coarse, multinucleate, and nonseptate.

Eriksson's mycoplasm hypothesis gained little support, but he reiterated his faith in it in 1905 (*18*) and again in 1921 (*19*).

Much of the later cytological work on stripe rust centered on the question of mycoplasm. In 1900 Klebahn (*28*) noted that the stripe-rust hyphae are few and coarse and multinucleate and can be traced along the length of the leaf for considerable distances. Near a uredinium the hyphae have more cross walls and fewer nuclei. He figured and described the haustoria and surmised that they were the "special corpuscles" or "Plasmanucleoli" forming the basis of Eriksson's mycoplasm hypothesis. In 1904 Klebahn (*29*) published figures of stripe rust showing host cells filled with "dickem Plasma" and containing numerous fungous nuclei and also a rust hypha containing a host nucleus. He regarded these, however, as artifacts ("Kunstproducte") and continued skeptical of the mycoplasm theory.

Ward, in England, was an able antagonist of the mycoplasm hypothesis. In 1903 (*37*) he published a cytological study of the uredinial generation, using principally *Puccinia dispersa*, and traced the development through from the germination of the spore. Ward contended, and correctly, that Eriksson's "corpuscles spéciaux" were haustoria and that Eriksson reversed the order of events in stating that they form in the host cell from the hypothetical mycoplasm and then move out into intercellular spaces to form hyphae.

In 1905 Ward (*38*) again discussed mycoplasm, reporting experiments to show the great longevity of urediniospores, their broad distribution by wind, the conditions governing their germination, and the probability of their overwintering. He described the germ tube, the multinucleate substomatal vesicle, and the large, multinucleate, nonseptate hyphae of *Puccinia glumarum*. He found no evidence of

the procedure described by Eriksson. The appearances of *P. glumarum* on a resistant wheat and on a susceptible wheat deprived of carbon dioxide are similar. In both, the hyphae soon become smaller and starved looking, their nuclei become indistinct, haustoria are rare, and host cells collapse and stain red.

Marryat (*31*) in 1907 studied cytologically the development of *Puccinia glumarum* on susceptible and immune varieties of wheat. On the susceptible host the germ tube enters without forming an appressorium. The vesicle has two nuclei and the stout infecting hypha four. Later hyphae are large and multinucleate, and haustoria are small and globular. About the tenth day "the nuclei now arrange themselves in pairs along the length of the hyphae which become divided up by septa." Spores are binucleate and are produced in abundance. On a resistant host entry is similar, but the contents of the hyphae soon become watery and then die, staining deep red. Haustoria are rare and minute, and host tissues die.

Pole Evans (*21*) in 1907, working in Ward's laboratory, found in *Puccinia glumarum* a cylindrical, multinucleate, substomatal vesicle which forms at one end a single infecting hypha. The appressorium was mentioned but not figured. The short branches of the mycelium are frequently septate, and the long branches are vacuolated, rarely septate, and apparently do not form haustoria. Long hyphae will grow 12 mm., then branch at the apex and start a new uredinium. Haustoria may be simple or branched and often contain as many as five nuclei. After a haustorium has formed from the tip of a hypha, a transverse septum makes its appearance and cuts off the tip. Later, many nuclei of the hyphae degenerate, and hyphae become very thin. Septation occurs the day before spore formation.

Lindfors (*30*) in 1924 studied cytologically plants bearing the first outbreak of rust in the spring and also later infections. The two were essentially alike; he found no evidence of mycoplasm. The binucleate spore forms a germ tube with two nuclei. The vesicle is thick walled and pushes out divergent infecting hyphae. Later, hyphae shape themselves to the space they occupy and are thin walled and multinucleate. Still later, long runners develop with septum formation at the branches. Then follows septation into binucleate cells, accompanied by degeneration of some of the nuclei. As reproduction begins, the nuclei of the mycelium become faint and the cytoplasm scant. The mycelium is to be considered as binucleate in principle because it returns to the binucleate condition during reproduction.

From the foregoing, it is evident that the cytological studies of stripe rust have been fragmentary and frequently at variance.

Eriksson's experiment of growing rusted plants from rusted seed in closed glass cages excited widespread interest. The experiments were repeated in Germany by Klebahn (*27*), in England by Massee (*32*), in Austria by Zukal (*40*), and in America by Bolley (*11*), and later by Hungerford (*24*), with uniformly negative results. The plants from rusted seed, grown in rust-proof cages, remained rust free.

Beauverie (*6*) in 1914 found that spores of *Puccinia glumarum* in seeds remained viable until February, and he considered it possible that the rust overwintered by this means.

During this time, however, evidence was accumulating throughout Europe that stripe rust can overwinter as hibernating mycelium in

leaves of grain and grasses. Spore production stops during cold weather, but, if the host survives, the mycelium produces fresh spores in the spring. This evidence was compiled by Henning in 1919 (*22*).

Attempts have been made to determine differences in chemical composition between varieties of wheat resistant and those susceptible to stripe rust. Henning and Bygden (*22*) in 1919 determined the acid and sugar content of several wheat varieties. No correlation was observed between either acid or sugar content and resistance. The most susceptible varieties had an exceptionally high sugar content, but so did Thule wheat, which was resistant. Arrhenius (*5*) in 1924 found no connection between the titration acidity and resistance to yellow rust.

Biffen (*9, 10*) made crosses between varieties of wheat immune from yellow rust and varieties susceptible to it. The F_1 generation was susceptible. The F_2 consisted of three susceptible to one immune. The immune plants bred true in succeeding generations. The others presented considerable range in susceptibility, but not more than would be accounted for by differences in manuring. In the F_3 they showed again a 3-to-1 ratio. Rust resistance was considered due to a single factor, inherited independently of all the morphological characters studied. Brooks (*12*) in 1921 and Armstrong (*4*) in 1922 were in substantial agreement with this.

Nilsson-Ehle (*33*), on the contrary, found transgressive segregation when varieties of different rust resistance were crossed and even when varieties of equal rust resistance were crossed, and he believed that several independent factors were concerned in producing the results. Pesola (*34*) also found evidence of more than one factor, and suggested that the varying results of investigators in different countries might be due to their working with different forms of the rust. Substantial progress in breeding rust-resistant varieties has been made by Biffen in England, Nilsson-Ehle in Sweden, Pesola in Finland, and others.

MATERIAL AND METHODS

Spores of stripe rust and seed of Jenkin club wheat were obtained from C. W. Hungerford, of Moscow, Idaho. Plants were grown and infected, and material fixed.

Later, stripe rust was found growing on *Bromus marginatus* Nees (E. 25, T. O. 780) in the grass garden in Berkeley, Calif., in the summer of 1926. The stripe rust was dormant, although another rust on the same plants was growing actively. When the fall rains came the stripe rust renewed its growth and the fresh spores were transferred to seedlings of *B. marginatus*. Transfer to several varieties of wheat showed that it made normal growth. The rust probably was *Puccinia glumarum tritici*.

In inoculating, rain water was used for spraying the plants. Neither tap water nor distilled water produced good results. The plants were kept in a moist chamber for 48 hours after inoculation, then placed under cheesecloth cages in greenhouse and field. The highest greenhouse temperatures of summer (90°–105° F.) proved fatal to the rust, although the hosts survived.

Bromus marginatus has proved a favorable host for culturing the rust, as the individual leaves are long lived, and a single infection can live and continue growth for five, six, and even seven weeks. Chains of uredinia reach a length of 10 and 11 cm. If the plants are kept well watered, fresh spores are always available.

In the spring of 1926, stripe rust appeared in the wheat plots at Davis, Calif. Material was fixed on April 22. At this time several rows in the identification nursery were heavily infected, and scattered infections had appeared at a number of other points in the field. Material was fixed from row 162 (C. I. 7554, a wheat from Scotland), row 163 (C. I. 7555–1, a wheat from India), row 165 (like 163), row 166 (C. I. 7556, a wheat from Algeria), and White Federation (C. I. 4981). Later on, May 14, 1926, telia on these same hosts were fixed. It was interesting to observe that although the rust had been growing and spreading vigorously when seen in April, it had made no further headway by the middle of May. The hotter, drier weather of May had checked its spread. Other wheat rusts, on the contrary, were increasing rapidly at the later date. Beauverie (7) made similar observations on stripe rust in France. The temperature range of stripe rust appears to be distinctly lower than that of the other wheat rusts.

Material was fixed in the chrom-acetic-urea mixtures and in Flemming's medium and weak solutions. It was washed and dehydrated by the usual methods, embedded in 50° paraffin, and the sections were cut 10μ thick and were usually stained in the triple stain. Hanging drop cultures of germinating spores were fixed over fumes of osmic acid, stained, and mounted in balsam.

INVESTIGATIONS

The urediniospores of *Puccinia glumarum* are bright yellow in color. Under the microscope the color is found to be in the cell contents. The wall is colorless. When mounted dry the spore wall is seen to be minutely echinulate. The average size is 22.5μ by 17.4μ. In water, the spores swell in a few seconds, becoming more nearly globular as they enlarge, averaging 29.5μ by 27.1μ. The stretched walls are thinner, of course, and are under considerable tension. Under slight mechanical pressure the spores burst and the empty walls contract, but not quite to the original size, at least not while remaining in water. They now average 24.6μ by 19.8μ.

In the spore wall are scattered germ pores. They range from 10 to 16 in a spore, and the average of 20 is 13.5. These are obscure in the living material but are readily brought out by staining. They are clearest in germinated spores whose contents have passed out into the germ tube. (Pl. 1, A, B, C.) In a preparation stained with gentian violet (pl. 1, B) the germ pores appear as small lighter-stained circles. When stained with safranin (pl. 1, C, *a*, *b*) each pore has a darker staining center. In edge view (pl. 1, D, *a*, *b*, *c*) the germ pores appear as thickened pads of light-staining material.

The yellow color of the spore contents is lost in fixed and sectioned material, and if the spore wall has not been too heavily stained the contents of the spore can be made out. (Pl. 1, D.) These consist of rich cytoplasm and usually two nuclei.

SPORE GERMINATION

Under favorable conditions the spores germinate in a few hours. On the leaf, where moisture conditions are not uniform, some spores may germinate at once, others not until several days after inoculation. The germ tubes follow closely the irregularities of the leaf surface, and in sectioned material it is seldom possible to follow a single germ tube for any distance. The growing tip of such a tube is seen in

Plate 1, E. It is uneven in thickness and somewhat swollen near the tip, *b*, where two nuclei lie close together.

In a hanging drop culture 1 day old, fixed and stained, the process of germination can be traced. An early stage is drawn in Plate 1, A. From one of the germ pores, *a*, the tube, *b*, issued. It is filled with dense, alveolar cytoplasm throughout its length. No nuclei are visible; they probably are still within the spore wall. A slightly longer germ tube is represented in Plate 1, B. The spore and the proximal part of the germ tube are nearly empty. At the growing tip, *b*, the cytoplasm is dense, and back of it at *a* is a pair of nuclei.

The great majority of the germ tubes in one-day hanging drop cultures are much longer than this. The average of 10 which were measured is 980μ. The maximum is 1,428μ. In these longer tubes the basal portion (pl. 1, C) is empty, and practically all of the protoplasm is concentrated at the tip (pl. 1, F, G). Usually only one pair of nuclei is to be seen (pl. 1, F, *a*), but more rarely two pairs occur (pl. 1, G, *a*, *b*).

On the leaf, the germ tubes grow directly to the stomata. No well-marked appressorium, such as occurs regularly in the stem and leaf rusts of wheat, has been seen in stripe rust. On reaching the stoma, the germ tube swells slightly. (Pl. 2, A, *a*.) This occurs in material fixed one day after inoculation. The cytoplasm is dense at the tip and thins out gradually farther back, as would be the case in the germ tube before it reached the stoma. No septum has formed to isolate a terminal cell.

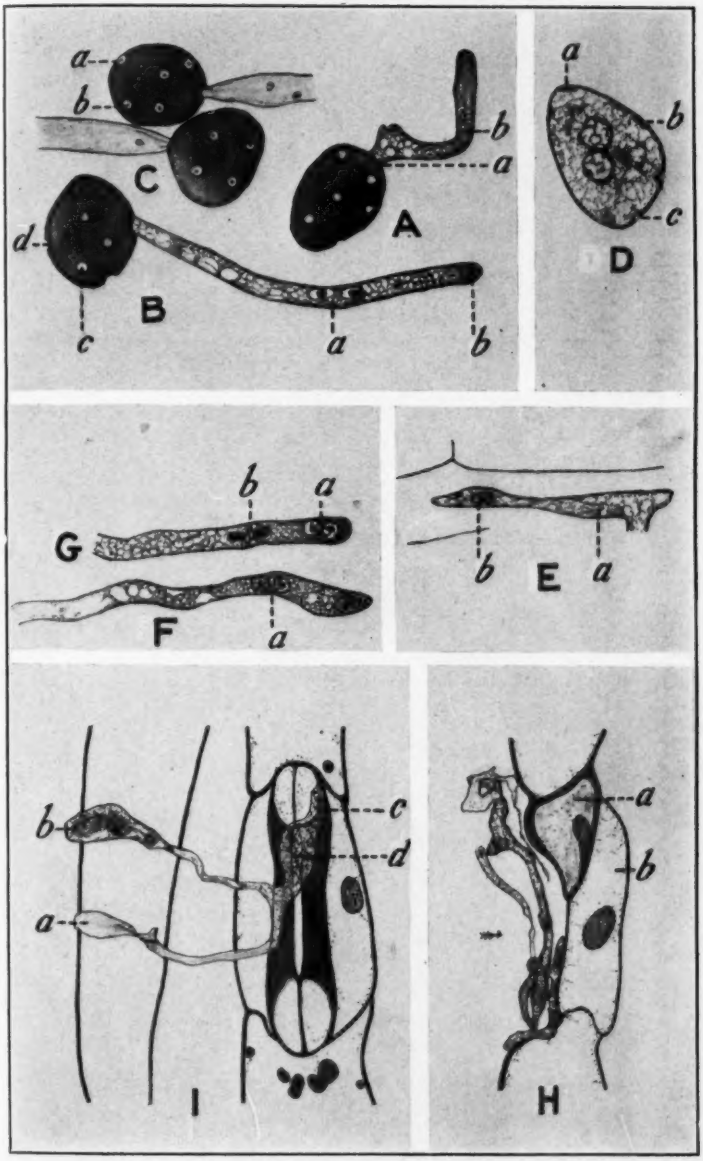
ENTRANCE OF THE FUNGUS

Entry of the fungus is usually very prompt, but even when it is delayed the fungous plasm does not round up into a cushion over the stoma. In Plate 1, H, from material fixed three days after inoculation, is represented a somewhat oblique section through a stoma, cutting through the end of a guard cell at *a*, and an accessory cell of the stoma at *b*. Several germ tubes have reached the stoma and, having failed to enter, have twisted about on its surface. No true appressoria have formed.

In a previous study of leaf rust (*Puccinia triticina*) (*3*), it was found that in a small percentage of cases where several germ tubes reached the same stoma the appressoria formed there might fuse, or might enter separately and fuse after entry. No conclusive evidence of a similar process has been seen in stripe rust, although some of the material suggests it. In Plate 1, I, is shown a surface view of a stoma on which three germ tubes, *a*, *b*, and *c*, converged at *d* where entry is in progress. The central fungous mass at *d* is apparently continuous, but the dark-stained guard cell walls underneath make it difficult to be sure of this. If fusion occurs it is rare, in this material

EXPLANATORY LEGEND FOR PLATE 1

A.—Hanging drop culture. The spore, *a*, has formed a short germ tube, *b*. Germ pores distinct. × 730.
B.—Later stage of germination. The germ pores, *c*, *d*, show clearly on the empty spore. The cytoplasm of the germ tube is densest at the tip, *b*. Two nuclei at *a*. × 730.
C.—Still later. Spore and basal part of tube empty. Germ pores at *a* and *b*. × 730.
D.—Spore showing contents—two nuclei and cytoplasm. Germ pores, *a*, *b*, *c*, distinct. × 1130.
E.—Tip of germ tube on leaf showing pair of nuclei, *b*, and vacuolate cytoplasm farther back at *a*. × 730.
F.—Tip of old germ tube from hanging drop culture. Two nuclei at *a*. Basal part nearly empty. × 730.
G.—Similar. Two pairs of nuclei, *a* and *b*. × 730.
H.—Three days after inoculation of *Bromus marginatus*. Germ tubes reached stoma but failed to enter. Portion of guard cell at *a*, and accessory cell at *b*. No appressoria formed. × 730.
I.—Four days after inoculation on *B. marginatus*. Three germ tubes, *a*, *b*, *c*, converged at the stoma at *d* and are entering, apparently as one mass. × 730.



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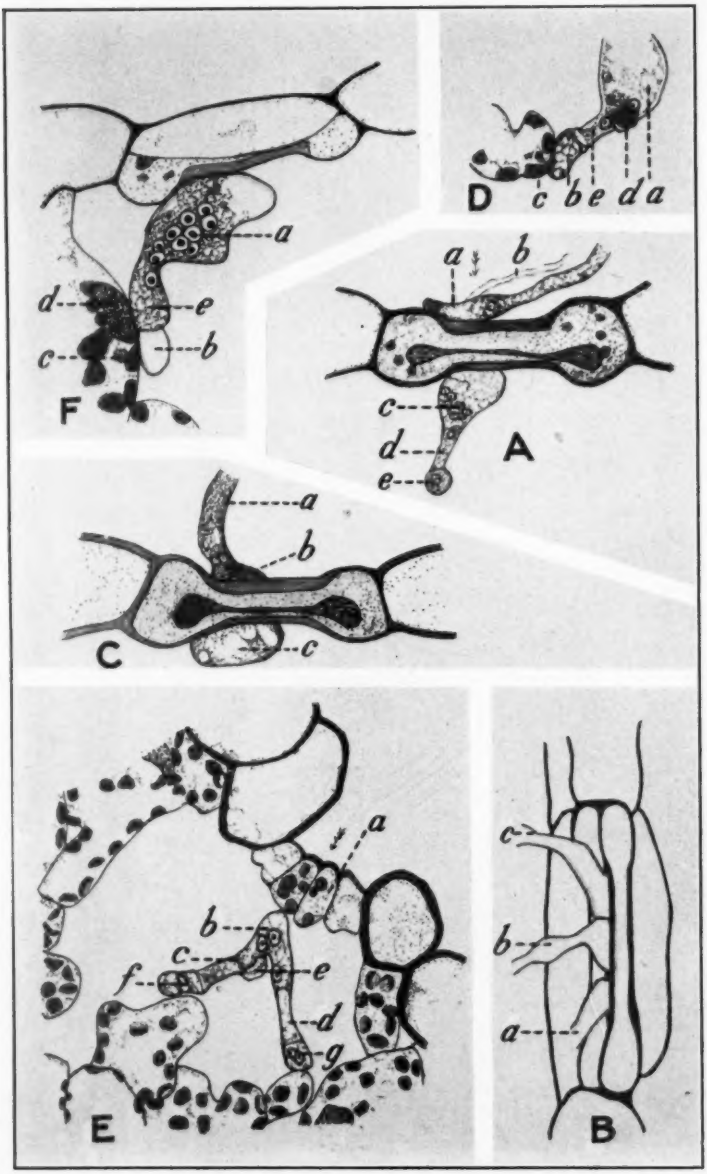

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at least. Ordinarily when several germ tubes reach the same stoma they remain separate. In Plate 2, B (drawn in outline only), the three germ tubes, *a*, *b*, *c*, are distinct.

Entry takes place immediately if conditions permit. There is ordinarily no pause at the stoma. In Plate 2, C, drawn from material fixed only 16 hours after inoculation, entry has begun. It usually takes place, as in this case, at one end of the stomatal slit. The germ tube reached the stoma at *b*, spread slightly along the crack between the guard cells (only one of which is drawn), pushed through, and is swelling out into a rounded vesicle at *c*, on the inner surface of the stoma. The bulk of the protoplasm is still outside in the germ tube, *a*, *b*, where the contents are relatively dense.

In the example just described, the germ tube spreads somewhat along the stoma before entering, but this is not essential. The unmodified tip of the germ tube can enter directly, merely becoming somewhat flattened as it passes through the narrow slit.

Here, too, no septum can be seen in the germ tube. (Pl. 2, C.) It may be just this lack of a delimiting wall which prevents the formation of the large rounded appressorial cushion typical of other rusts. A little later, however, when entry is completed, a septum occasionally may be seen. (Pl. 4, A, *f*.) It is not uniformly present.

FORMATION OF INITIAL HYPHA AND HAUSTORIUM

Plate 2, A, is drawn from material fixed 24 hours after inoculation. Two germ tubes reached this stoma. The one, *a*, already mentioned, came last. Another arrived earlier and has entered. An empty withered germ tube, *b*, is all that remains of it outside the stoma. Inside are the substomatal vesicle, *c*, and a short hypha, *d*, swollen at the tip, *e*, in preparation for the formation of a haustorium. The cytoplasm is rather thin and the turgor low. Nuclear divisions have evidently occurred, for there are now 10 or 12 small nuclei.

In the case just described, a single hypha formed from the substomatal vesicle. More commonly there are two or three. As they extend in divergent directions, only a fraction of the whole occurs in any one section. In Plate 2, D, drawn also from one-day material, the partly evacuated vesicle, *a*, gave rise to two hyphae, one at *e*, which has formed a haustorium mother cell, *b*, and the beginning of a haustorium at *c*. The base of a second hypha is at *d*, the rest having been cut off in sectioning.

Another view of this stage is drawn in Plate 2, E, which is a detail from a cross section of a leaf fixed 24 hours after inoculation. Underneath the cross section of the stoma, *a*, is the large substomatal air chamber bounded by mesophyll tissue. In this air space is the end

EXPLANATORY LEGEND FOR PLATE 2

Infections on *Bromus marginatus*

A.—One-day infection. One germ tube, *a*, has just reached the stoma. The second, *b*, is empty, the contents having entered to form the substomatal vesicle, *c*, which in turn has formed a short infection hypha, *d*, terminated by a haustorium mother cell, *e*. × 730.

B.—Sixteen hours after inoculation. Three germ tubes, *a*, *b*, *c*, (outline only) reached the same stoma. No appressoria formed, and there is no fusion. × 730.

C.—Early stage of entry. The germ tube, *a*, reached the stoma at *b*, and is passing through to form the vesicle, *c*. × 730.

D.—One-day infection. The substomatal vesicle, *a*, pushed out two hyphae; one at *d*, sectioned, and a second, *e*, which has formed the haustorium mother cell, *b*, and the beginning of a haustorium *c*. × 730.

E.—One-day infection. Cross section showing stoma, *a*, end of vesicle, *b*, two infecting hyphae, *c* and *d*, terminating in haustorium mother cells, *f* and *g*. A third infecting hypha, *e*, was cut in sectioning. × 730.

F.—Two-day infection. Enlarging substomatal vesicle at *a*. Empty haustorium mother cell, *b*, and end of haustorium, *c*, accompanied by host nucleus, *d*. Hypha swelling at *e*. × 730.

of the substomatal vesicle, *b* (the body of which is in an adjoining section), and two hyphae, *c* and *d*, each terminating in a haustorium mother cell at *f* and *g*, respectively. At *e* is the stub of a third infecting hypha, cut off in sectioning. Each of the haustorium mother cells is delimited by a septum and contains cytoplasm and two small nuclei. No other septa are to be seen. The number of nuclei in the first-formed haustorium mother cell is not uniform. Three and even four have been found, although two are most common.

The substomatal vesicles are small at this stage and meager in content. The average size of 10 is 17.5μ by 12.3μ. The longer diameter is often at right angles to the epidermis, as in Plate 2, A and D.

Under winter conditions (this material was infected in January) the rust spreads little if at all on the second, third, and fourth days after inoculation. It still is limited to the edges of the air space beneath the stoma of entry. Development during this time consists in the absorption of food from the host, an increase in cytoplasm, and multiplication of nuclei accompanied by a swelling of the vesicle and hyphae to several times their original dimensions.

The steps in this process can be followed. Two-day material presents considerable range in development. One of the early stages is shown in Plate 2, F. It is drawn on the same scale as the younger vesicles and hyphae in Plate 2, A and D. On comparing the two, the trend of development is at once apparent. The stage in Plate 2, A, has been attained without food other than that originally stored in the spore from which it grew. In Plate 2, F, there is an empty haustorium mother cell at *b* and the tip of a haustorium at *c*. The remainder of the haustorium lies in the next section. Once the food and water supplies of the host have been tapped, there is immediate expansion. Both vesicle and hypha are rounding out. The nuclei have not increased in number but are full grown.

Food absorbed from the host cell by the haustorium and passed on out through the mother cell is naturally most abundant just back of this cell. Here fungous plasm accumulates. There is formed regularly a rounded, almost globular swelling of the hypha at this point. The beginning of this swelling is seen at *e* in Plate 2, F. It is more pronounced at *a* in Plate 3, A, just back of the mother cell *b*. In Plate 3, B_1 and B_2, are drawn successive sections of the same fungus, which has formed two haustoria. Just outside of the host cell containing the haustorium, *g*, is the beaklike haustorium mother cell, *f*, and just back of it is the rounded swelling of the hypha *e*. In Plate 3, B_2, is the haustorium, *d*, connected with the mother cell, *c*, and back of it a large globular fungous mass, *b*. This connects with the part labeled *b* in B_1. The point at *a* connects directly with

EXPLANATORY LEGEND FOR PLATE 3

Infections on *Bromus marginatus*

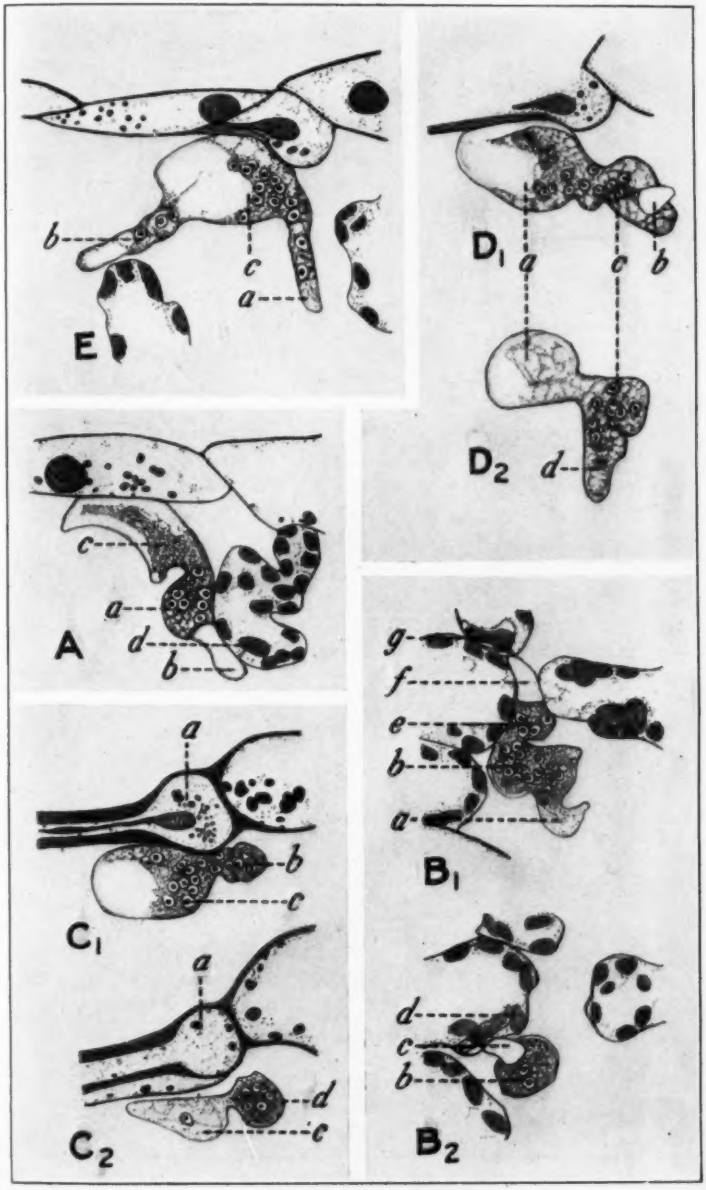
A.—Two-day infection with partly collapsed substomatal vesicle, *c*, and infecting hypha, *a*, greatly swollen just behind the haustorium mother cell, *b*. Haustorium at *d*. × 730.

B_1 and B_2.—Successive sections of a two-day infection. B_1, *a*, adjoins the substomatal vesicle (not drawn). There are two infecting hyphae, greatly, enlarged. One at B_2, *b*, with empty haustorium mother cell at *c* and haustorium at *d*. The second, B_1, *e*, with mother cell at *f* and haustorium, *g*. × 730.

C_1 and C_2.—Two-day infection. Successive sections of substomatal vesicle showing increase in size and in contents. Guard cell at *a*, substomatal vesicle, *c*, swollen infecting hypha, *b*, *d*. × 730.

D_1 and D_2.—Four-day infection showing the substomatal vesicle, *a*, the empty haustorium mother cell, *b*, the hypha greatly swollen just back of it at *c*, and the growing tip, *d*. No septa except at *b*. × 730.

E.—Four-day infection with the substomatal vesicle, *c*, forming an infecting hypha at each end, *a* and *b*. × 730.



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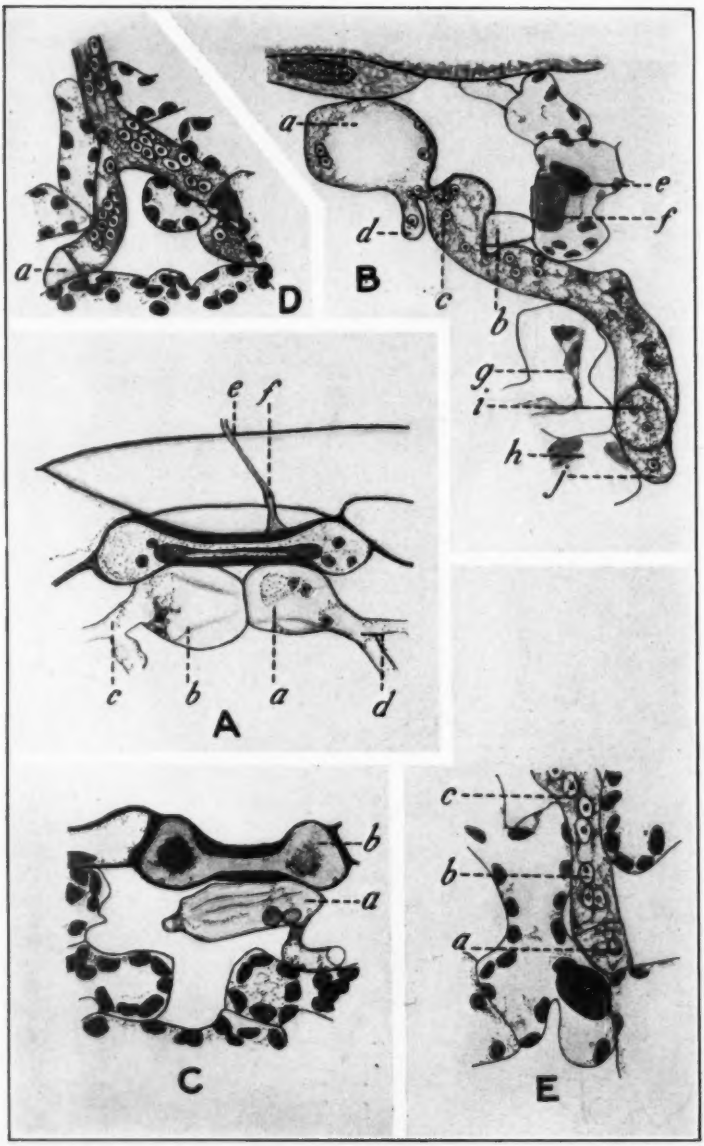

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the substomatal vesicle. There are over 2 dozen nuclei in this irregularly bulging mass. There are only two septa—the ones delimiting the two haustorium mother cells.

One result of the absence of cross walls is a greater freedom of motion. There are no nuclei in the short hypha at *d* in Plate 2, A, nor at *e* in Plate 2, F. A little later all of the nuclei are at the corresponding position, *a*, in Plate 3, A.

When the substomatal vesicle first rounds out in this process of expansion it is rather unstable and sometimes collapses under the action of the fixing fluid. (Pl. 3, A, *c*.) A little later it holds its form. Moreover, as the protoplasm grows and the swelling hypha is densely filled, the protoplasm often backs up into the vesicle, for there are no cross walls to stop it. In Plate 3, C_1 and C_2, still from two-day material, the vesicle, *c*, is more than half full of cytoplasm rich in nuclei. It is several times its original size (cf. pl. 2, A and D) and has changed shape, for the long diameter is now regularly parallel to the long axis of the guard cells. Ten substomatal vesicles from three-day material have been measured. The average size is 26.8μ by 17.1μ, which contrasts sharply with the original size (17.5μ by 12.3μ).

Similar figures still may be found on the fourth day. In Plate 3, D_1 and D_2, successive sections of the same organism, there is the half-filled vesicle, *a*, against the guard cell. The haustorium mother cell, *b*, originally the tip of a slender hypha, looks now like a little lateral appendage on a short massive hypha whose apex is at *d*.

Although two or three hyphae may form from the same vesicle, they usually issue near each other from the same end of the vesicle. (Pl. 2, D and E.) Occasionally, however, one finds hyphae coming from opposite ends of the vesicle, as in Plate 3, E, *a* and *b*. So far as noted, the development of the two in such cases is unequal. As in this instance, the bulk of the cytoplasm in the vesicle is at one end, continuous with the hypha, *a*, which in turn connects (in adjoining sections not drawn) with the usual haustorium mother cells (two in this case) and globular swellings. There are over 40 nuclei in this fungus, of which only 3 occur in the hypha at *b*. The further development of the latter is doubtful.

Occasionally two germ tubes reach the same stoma and both enter. Figures like the one drawn in Plate 4, A, are not uncommon. At *e* is the empty germ tube leading to the vesicle, *a*. The survival of the germ tube to this stage (fourth day) is rare. It usually disappears soon after entry; in fact, it is rarely seen even on the second day. The two vesicles, *a* and *b*, are quite distinct, and each forms its own infecting hyphae, *c* and *d*. In this case the vesicles are nearly emptied, the cytoplasm and nuclei having passed out along the lines of growth.

EXPLANATORY LEGEND FOR PLATE 4

A.—A four-day infection on *Bromus marginatus*. Two entries occurred at the same stoma. The two nearly empty substomatal vesicles, *a* and *b*, are not joined. Each produced two infecting hyphae, *c* and *d*. Remnant of germ tube at *e*, *f*. × 730.

B.—From a seven-day infection on *B. marginatus*. The substomatal vesicle, *a*, formed two hyphae; one abortive, *d*, the other forming a mother cell at *b* with dead haustorium, *f*, accompanied by dead host nucleus, *e*. The hypha expanded at *c*, and formed the nonseptate hypha, *c*, *j*, with branch at *i*. Dead host cells at *g*, *h*. Multinucleate, nonseptate mycelium from this extends 450 μ. × 730.

C.—Seven-day infection on *B. marginatus* with empty withered substomatal vesicle at *a* beneath dead guard cell, *b*. × 730.

D.—Seven-day infection on *B. marginatus*. Branching hypha in mesophyll. Nuclei irregularly arranged. No septa except those delimiting the haustorium mother cells, as at *a*. × 730.

E.—From 17-day infection on Jenkin club wheat. Hypha, *b c*, terminates in a haustorium mother cell, *a*. × 1130.

The time when this evacuation of the vesicle occurs varies greatly. In Plate 4, A, four days after inoculation, it is nearly complete. In Plate 4, B, on the contrary, seven days after inoculation, cytoplasm and nuclei are still present, and the vesicle has retained its turgor. The evacuation of the vesicle is accompanied by its shrinkage. This is beginning in the vesicles of Plate 4, A, and is much more pronounced in later preparations. In Plate 4, C, seventh day, the vesicle is small and wrinkled, and contains mere traces of cytoplasm and nuclei.

The material used in this study of early stages of infection was grown in the field in January, and the question arose as to whether winter conditions, particularly low temperature, had retarded the growth. For comparison, a second lot of inoculations was made in April. The early stages resembled closely those of the winter material, both as to method and rate of development. All evidence available at present indicates that this prolonged juvenile period, during which the organism increases in thickness without adding materially to its length, is normal to this species.

DEVELOPMENT OF THE MYCELIUM

After this long preliminary period, during which the primary hypha or hyphae expand slowly to the diameter of later stripe-rust hyphae, a hypha grows out into the mesophyll tissue, working its way through the irregular intercellular spaces. The direction of growth is at first indifferent. It may grow lengthwise of the leaf, but it is quite as apt to grow transversely or diagonally.

In Plate 4, B, representing part of a seven-day infection, the substomatal vesicle at *a*, already mentioned, is living. Two hyphae formed from it, one at *d*, abortive, and a second which originally terminated in a haustorium mother cell at *b*. The haustorium from it, *f*, is now dead, and alongside it is the host nucleus, *e*, also dead. The haustorium evidently functioned, however, for the typical rounded swelling, *c*, formed just back of the mother cell, and from it grew the hypha, *c-j*. A branch at *i* (cut in sectioning) leads out into deeper tissues, where it has branched and spread lengthwise of the leaf through a distance of 450μ. The entire mycelium is without septa other than those delimiting haustorium mother cells. This absence of barriers facilitates the transfer of materials along the hyphae and may explain why this hypha, *c-j*, can still contain rich protoplasm, although the host cells around it, *g*, *h*, *e*, are dead or dying.

Something of the habits of the mycelium may be seen from Plate 5, A, drawn from a six-day preparation. Coming in at *b*, the hypha passes under a mesophyll cell, reappears at *d*, grows down to *k*, nar-

EXPLANATORY LEGEND FOR PLATE 5

A.—Six-day infection on *Bromus marginatus*. Branching hypha, *b*, *k*, *l*, *m*, in mesophyll. Haustorium mother cells at *a*, *c*, *d*, *f*, and *i*. Haustoria at *e*, *g*, and *j*. Host-cell nucleus at *h*. No septa except for haustorium formation. × 730.

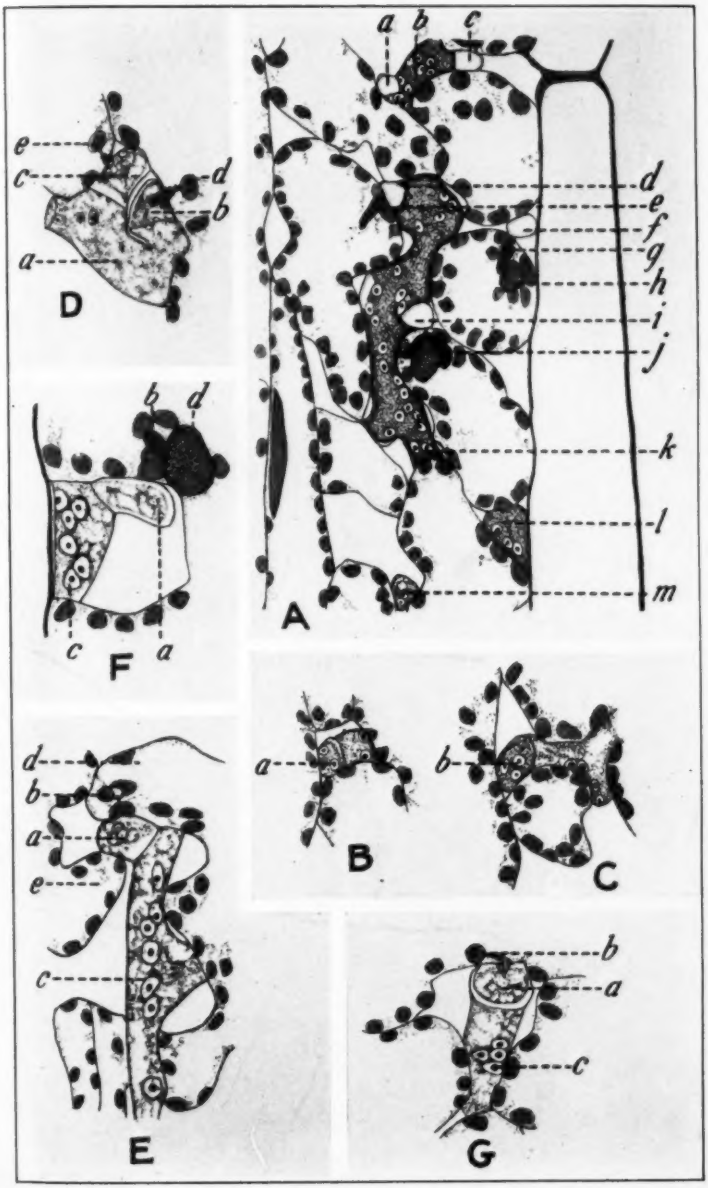
B and C.—From six-day infection of *B. marginatus*. Newly formed haustorium mother cells at *a*, *b*, one with two nuclei, the other with three. × 730.

D.—From 17-day infection on Jenkin club wheat. Two haustorium mother cells, *b* and *c*, formed on the hypha, *a*, are beginning to form haustoria at *d* and *e*. The necks of these haustoria are encrusted with granular matter. The nuclei in the mother cells are indistinct. × 1130.

E.—Seventeen-day infection of Jenkin club wheat. The hypha, *c*, formed a mother cell, *a*, between the host cells, *d* and *e*. The mother cell is forming the haustorium, *b*. Nuclei of the mother cell are doubtful. × 1130.

F.—Similar to E. On the hypha, *c*, has formed a haustorium mother cell, *a*. No nuclei visible in the mother cell. Young haustorium at *b*, with host nucleus, *d*. × 1130.

G.—Similar to E. Hypha at *c*. Contents of mother cell, *a*, without visible nucleus. Young haustorium at *b*. × 1130.



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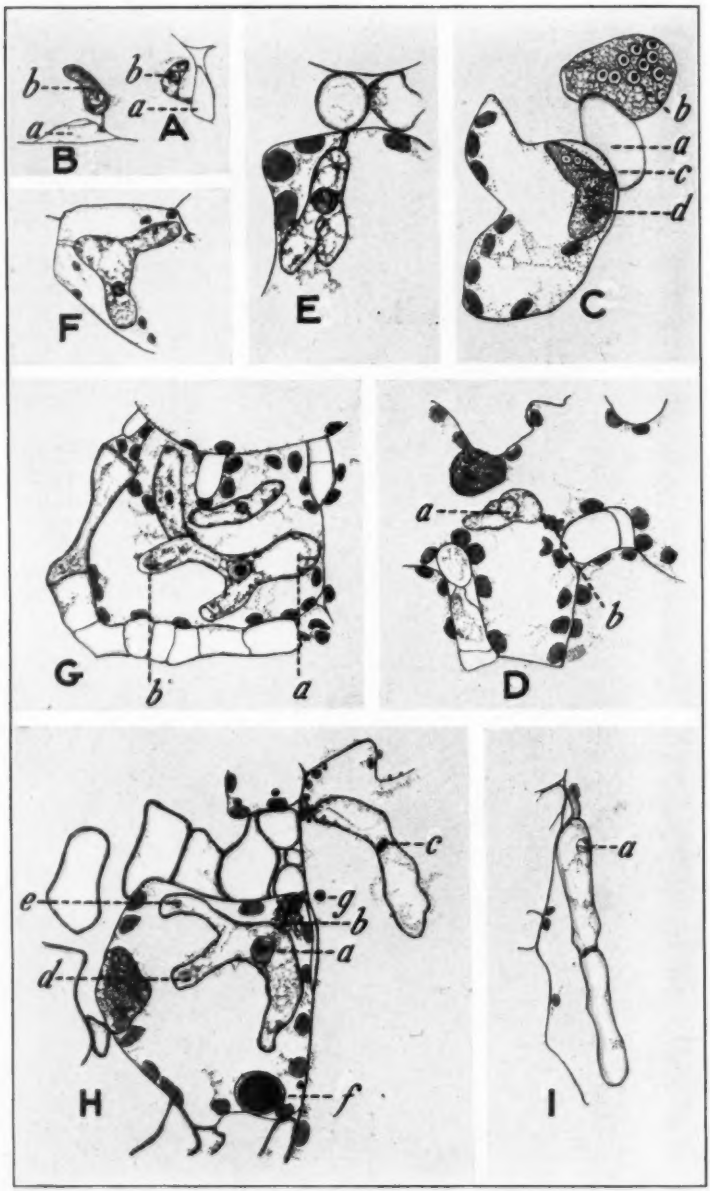

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rowing where the air space narrows, broadening out where there is room, disappearing at *k*, and coming again into the plane of the section at *l* and *m*. Numerous nuclei are scattered along its length in no regular order. So far as can be detected, the nuclei are not paired.

The characteristic haustorium mother cells occur at Plate 5, A, *a*, *c*, *d*, *f*, and *i*, each connected with its haustorium as at *e*, *j*, and *g*. In *j* and *g* the host nucleus lies in contact with the haustorium. The tendency to form an expanded mass of fungous plasm in the hypha just back of the haustorium mother cell, where food is coming in, is less pronounced but can still be seen in some cases. In the hypha just back of the mother cell, *d*, is a large rounded mass almost filling the large air space. Food absorbed by the haustorium, *j*, and passed out through the mother cell, *i*, has brought about a local expansion of the hypha at that point, but space limitations have forced the swelling hypha around the mother cell itself, almost submerging it.

In larger air spaces the hyphae take a cylindrical form (pl. 4, D) and are far more uniform in thickness. The average diameter is about 9μ. Here, too, the fungous nuclei are distributed in irregular order, often with 10 or 12 in a group. There is no obvious pairing.

Haustoria form in the same fashion in mycelia of all ages, but older infections have proved most favorable for a detailed study of all stages of the process. For some reason not understood, haustoria of young infections often collapse in the fixing fluid even when the rest of the fungus and the host tissues are excellently fixed. In older infections some difference, probably in degree of turgor, resulted in much better fixation of haustoria.

When the tip of one of these coarse multinucleate hyphae forms contact with a host cell or becomes wedged into an angle of some small intercellular space, a septum forms a short distance back from the tip, giving rise to a short broad terminal cell, the haustorium mother cell. (Pl. 5, B, *a*, and C, *b*.) The nuclei of this cell decrease rapidly and unequally in size and become very difficult to see. Only when the cell is first delimited can the nuclei be counted. In the first formed haustorium mother cells (pl. 2, E, *f* and *g*) the number of nuclei commonly is two. In Plate 5, B, *a*, there are two; in C, *b*, there are three. In other cases four, five, and even six have been counted. At a slightly later stage (pl. 4, E), although the numerous nuclei of the hypha, *b*, *c*, are clearly defined and conspicuous, those in the mother cell are small and vague. One little nucleus at *a* is fairly clear, but the smaller, fainter circles near it

EXPLANATORY LEGEND FOR PLATE 6

A.—From 17-day infection on Jenkin club wheat. Young haustorium beginning to expand with single nucleus at *b*. Empty mother cell at *a*. × 1130.

B.—Similar to A. Empty haustorium mother cell, *a*, and uninucleate haustorium, *b*. × 1130.

C.—From four-day infection on *Bromus marginatus*. Mother cell, *a*, with swollen hypha, *b*, behind it. The small haustorium, *c*, is binucleate and accompanied by the host nucleus, *d*. × 1130.

D.—From 17-day infection on Jenkin club wheat. Expanding haustorium, *a*, with encrusted neck, *b*. × 1130.

E.—From infection on leaf sheath of wheat, C. I. 7555–1. Half-grown, branched, uninucleate haustorium. × 1130.

F.—From infection on leaf blade of wheat C. I. 7554. Forked haustorium with divergent branches. × 1130.

G.—From infection on glume of wheat C. I. 7555–1. Several haustoria in one cell. Haustorium, *a*, *b*, slightly plasmolyzed at *b*. Host cell living. × 1130.

H.—From infection on leaf sheath of wheat C. I. 7555–1. Haustorium, *a*, with three branches slightly plasmolyzed at *d* and *e*, and heavily sheathed neck, *b*. Second haustorium at *c*. Intracellular bodies at *f* and *g*. × 1130.

I.—From infection on leaf sheath of wheat C. I. 7555–1. Large haustorium (39 μ long) almost completely drained. Single haustorial nucleus at *a*. × 1130.

may be either degenerating nuclei or merely heavier bits of cytoplasmic reticulum.

In Plate 5, E, the end of the hypha became firmly wedged between two host cells, *d* and *e*, and formed the mother cell, *a*. Within it are four small circles. Whether any or all of these are nuclei can not be determined. In this case, part of the content of this cell has moved in to the host cell, beginning the formation of a haustorium at *b*.

In Plate 5, F and G, the nuclei in the mother cells, *a*, *a*, either have become so faint or so small as to be indistinguishable or have already entered the haustoria, *b*, *b*.

In Plate 5, D, the hypha *a* has produced two mother cells, *b* and *c*, crowded into a small intercellular space and shaped closely to its outline. The one at *b* is forming the haustorium *d* and the one at *c* is entering another host cell at *e*. The necks of these young haustoria, *d* and *e*, are not slender like the others depicted, but are seemingly encrusted by granular matter. Whether this coating is derived from the host or from the rust is not known, nor is it known what conditions lead to its formation. It occurs more commonly in old than in young infections.

The very young haustorium is too dense and stains too deeply to show the nature of its contents. Apparently the protoplasm of the mother cell becomes concentrated as it flows through the narrow neck of the haustorium. It soon absorbs water from the host, however, and expands into a more open vacuolated structure. In Plate 6, A, at *b*, is a young haustorium only 4.5μ long, which shows clearly a single well-formed nucleus and reticulated cytoplasm. Its mother cell, *a*, is empty. The slightly older haustorium (pl. 6, B, *b*), 7μ long, also shows a single clearly defined nucleus. Whatever the number of nuclei in the mother cell may have been, but one reappears in the haustorium, and it expands with the growth of the haustorium. A few exceptions have been noted. The haustorium at *c*, in Plate 6, C, contains two small nuclei. The fate of the extra nuclei when a uninucleate haustorium arises from a binucleate or multinucleate mother cell has not been determined, but the fact that the nuclei decrease unequally in size before entering the haustorium suggests degeneration of the supernumerary nuclei.

The great majority of the haustoria develop into elongated unbranched bodies, thicker at the base and tapering somewhat toward the tip. The single nucleus lies about one-third of the way from the base. In Plate 6, D, the half-grown haustorium, *a*, is typical. Here again the neck, *b*, is encrusted with a blue-staining granular material of unknown origin. This occurs not infrequently in infections on a number of hosts, but there always are many haustoria with slender necks along with those that show this modification. It is doubtful whether it should be considered a sign of resistance in the host. Plate 6, D, was drawn from an infection on Jenkin club wheat, on which the rust makes good growth. It is found equally commonly on *Bromus marginatus*, a fully susceptible host; also on C. I. No. 7555–1, a susceptible wheat from India.

Occasionally a haustorium forks into two branches which may lie close together (pl. 6, E) or diverge (pl. 6, F and G, *a*). More rarely three branches are formed. (Pl. 6, H, *a*.) The single nucleus of these branching haustoria is usually stationed near the point of

origin of the branches. This last haustorium (pl. 6, H, *a*) has an unusually thickened neck, *b*, within which the slender original neck can be seen. Occasionally there is a slightly denser mass of cytoplasm near the tip of a branch of the haustorium (H, *d*, *e*). In case the haustorium is slightly plasmolyzed (pl. 6, G, *b*) this mass may bear a superficial resemblance to the haustorial nucleus. Usually, however, it is irregular enough in outline to be unmistakable.

Full-grown haustoria are 25μ or 30μ in length (counting each branch separately). The largest one measured (pl. 6, I) is 39μ long.

The host nucleus is often found associated with the haustorium. A count was made to determine the percentage of cases in which the haustorium and nucleus were together. Only those were counted in which the haustorium was full grown and in which both nucleus and haustorium could be seen clearly, and in mesophyll cells large enough so that there was room for them to have been widely separated. In 41 out of 50 cases, or 82 per cent, the two were together. This is too large a percentage to be accidental. Since many of these haustoria were located at the end of a cell three or four times as long as the haustorium, the contact between the two can have been achieved only by the motion of the nucleus to the haustorium.

Host nuclei in young and old infections and in uninfected tissue near by were measured. There is some expansion of the nucleus in infected tissue. The larger nuclei measured 10.0μ by 7.5μ in infected tissue and 8.7μ by 7.7μ in uninfected. The expansion is slight compared to that seen in infections of other rusts.

At the end of the first week of growth the mycelium has invaded an area from 350μ to 500μ in length, but not all of the intercellular spaces within that region are filled. The hyphae have not branched richly. Even at the center of the infection the fungous growth is still sparse.

Between the seventh and the ninth day the mycelium begins to spread rapidly through the leaf. This process is influenced markedly by the anatomy of the leaf.

In these sword-shaped leaves the veins run lengthwise of the leaf and parallel to one another. They are strands of vascular tissue circular in cross section and are embedded in rather dense mesophyll tissue with small intercellular spaces. From 25 to 50 per cent of the veins of later leaves are reinforced by heavy-walled strengthening tissue extending from the vein to both upper and lower epidermis. This tissue is practically without intercellular spaces. Each vein is marked on the upper side of the leaf by a ridge, so that this surface of the leaf is minutely corrugated. The stomata are arranged in longitudinal rows parallel to the veins. There usually is one row (occasionally two) on each slope between a ridge and its adjoining valley. The stomata are fewer on the lower surface of the leaf and are opposite those of the upper. Beneath each stoma is a large air chamber, and as the stomata form a close row (usually only one epidermal cell intervening between one stoma and the next), these rows of substomatal air chambers form chains of communicating passageways running from one end of the leaf to the other.

It is through these lanes that the fungus makes most rapid progress. Coarse, runnerlike hyphae traverse these air spaces, running as straight as the passageways in the leaf permit and following the lines of least resistance. (Pl. 7, A, *b*, *c*, *d*.) Even here, however, a runner

seldom grows more than 100μ, or 125μ at the most, without encountering a host cell. It may swerve and grow around it, but usually it stops, forms a haustorium mother cell and a haustorium (pl. 7, B, *a*, *b*, *c*), and continues growth by means of a branch pushed out just back of the haustorium mother cell (pl. 7, B, *d*, *e*). In young runners there are no septa other than those cutting off these short haustorium mother cells. In one case a runner was traced 400μ without a cross wall. The growing tip of a runner is dense and stains intensely. A little farther back (pl. 7, C) the contents are more open. The numerous nuclei are scattered or in groups with no obvious arrangement. The average diameter of these runners is 9.5μ. Still farther back from the growing tip (pl. 7, B) the runner, still nonseptate, shows scant content. There is a continuous central vacuole, and in the peripheral cytoplasm the nuclei are scattered or in pairs (pl. 7, B, *f*, *g*, *h*). Doubtless a part of the former content flowed on toward the growing tip, and some of it was utilized in the formation of lateral branches.

The rust also spreads transversely as far as continuous air passages in the mesophyll tissue permit. Here and there a runner gives off a small lateral branch which threads its way into the denser tissue, ramifying further and producing numerous haustoria. These feeding hyphae are variable in size, averaging 4.7μ in diameter. Many of the small intercellular spaces in the tissue around a vein are blind alleys, but some of them form continuous crooked passageways through which hyphae pass, and if the mycelium succeeds in reaching the more open tissue beyond the vein, a new set of runners is initiated, running parallel to the first. But the transverse spread of the mycelium is slow at best and is stopped on either side when a reinforced vein is reached, for the thick-walled strengthening tissue without intercellular spaces forms an impassable barrier.

So, while the transverse spread of the rust is checked when a reinforced vein is encountered, there is no mechanical barrier to the longitudinal growth, for the chains of substomatal passageways extend from one end of the leaf to the other. The result is a narrow band-shaped infection which can, and under favorable conditions does, extend the full length of the leaf.

When an infection is about 10 days old, a few septa form in the oldest part of the mycelium. These increase in number. As the mycelium extends longitudinally, the central septate area increases proportionately in size. In any older infection, all stages can be found between the nonseptate marginal region and the fully septate central area.

Some of the steps in this process have been observed. In Plate 7, D, is drawn a branching hypha in an early stage of septation. Septa,

EXPLANATORY LEGEND FOR PLATE 7

A.—From a 17-day infection of Jenkin club wheat. A longitudinal section showing partly septate runners, *b*, *c*, *d*, *f*, and finer mycelium in smaller air spaces, *g*, *h*, *i*. Subepidermal spore-producing hyphae massing at *a* and *e*. × 155.

B.—Detail from 17-day infection on Jenkin club wheat. Partly drained runner, *d e*, in substomatal air space. No septa except those delimiting the haustorium mother cells *a*, *b*, *c*. Nuclei paired at *f*, *g*, and *h*. × 730.

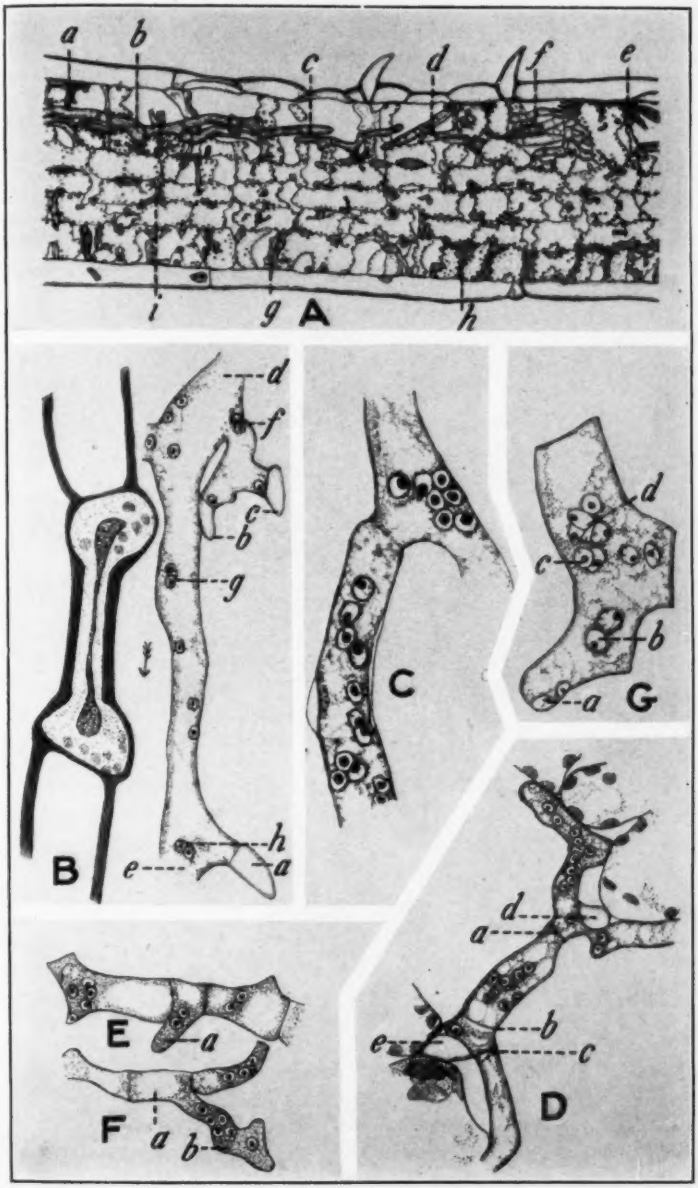
C.—Portion of nonseptate runner with rich cytoplasm and numerous irregularly arranged nuclei. × 1130.

D.—From 16-day infection on *Bromus marginatus*. Early stage of septation. Septa at *a*, *b*, *c*. Cells of hyphae still multinucleate. Empty haustorium mother cells at *d* and *e*. × 730.

E.—As in D. Showing passage of nuclei and cytoplasm of cell into the branch, *a*. × 730.

F.—As in D. The cell, *a*, nearly evacuated, its protoplasm having passed out into the branch, *b*. × 730.

G.—From 17-day infection on Jenkin club wheat. Detail showing paired nuclei at *a*, *b*, *c*, and larger group of nuclei at *d*. × 1130.



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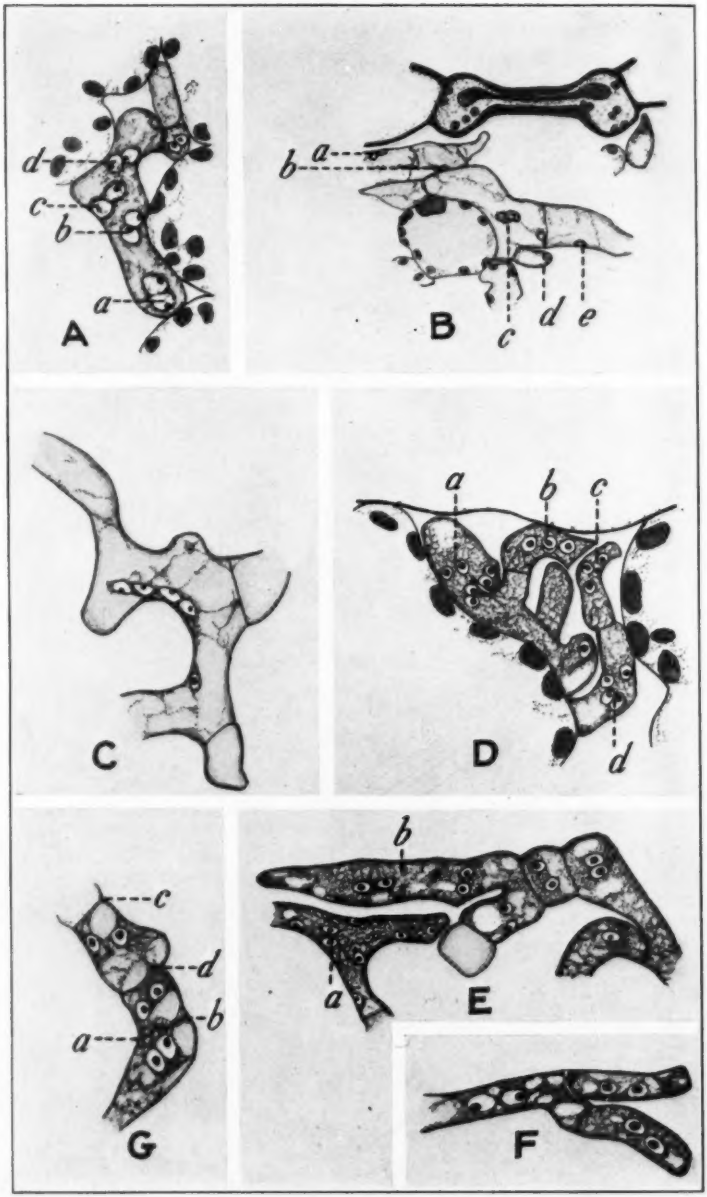

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of course, delimit the two empty haustorium mother cells, *d* and *e*, but in addition there are other septa, *a*, *b*, and *c*, cutting the main hypha into short lengths. These shorter cells are still multinucleate. The cell, *bc*, appears to be small and binucleate, but really extends on behind the mother cell, *e*. The nuclei are irregularly disposed in the cells, and the number in a cell varies.

These partly septate hyphae branch freely. In Plate 7, E, a binucleate cell has pushed out a short branch, *a*, into which both cytoplasm and nuclei are passing. In Plate 7, F, the cell, *a*, is nearly empty, the cytoplasm and six nuclei having moved out into the branch *b*. Sometimes a septum near the base of the branch cuts off retreat, and the evacuated portion of the parent hypha remains quite empty.

Septation progresses unevenly, varying within a small area. Alongside of hyphae with numerous irregularly disposed nuclei (pl. 7, C and D) are others in which some or all of the nuclei are arranged in pairs. In Plate 7, G, are three pairs of nuclei *a*, *b*, *c*, and an unassorted group, *d*. In Plate 8, A, is a hypha with four pairs of nuclei, *a*, *b*, *c*, *d*, and adjoining it a small cell which may be part of the same hypha, containing a fifth pair. In this case the two nuclei of a pair are close together, or even in contact, and the pairs tend to become scattered along the length of the hypha. Sometimes the pairing of nuclei is less obvious. Of course it is possible that these pairs are determined at an earlier stage, but repeated efforts to discover this have failed. Runners as well as the smaller hyphae become septate. Perhaps the pairing at *f*, *g*, and *h* in Plate 7, B, is a beginning of this process.

This rearrangement of nuclei normally is followed by the building of septa between one pair and the next, giving rise to binucleate cells. It does not work out perfectly, for an occasional cell, as in the runner figured in Plate 8, B, may have one pair, *c*, and an extra nucleus, *d*, while the adjoining cell has only one nucleus, *e*. Septation probably remains incomplete in places. In Plate 8, C, is drawn a hypha whose contents have been drained. The five nuclei are reduced in size, and the cytoplasm consists of a few delicate strands. Hyphae near by are septate, but it is doubtful whether the depleted living contents of this cell suffice for septum building.

The appearance of septa marks the beginning of reproductive activities. So regular is the sequence of septation and reproduction that the formation of septa in any part of an infection may be considered the first step in preparation for spore formation at that point.

DEVELOPMENT OF UREDINIA

Beginning at the center where the mycelium is oldest and best established, the wave of septation advances along the lines of growth,

EXPLANATORY LEGEND FOR PLATE 8

A.—From 17-day infection on Jenkin club wheat. Detail showing paired nuclei at *a*, *b*, *c*, *d*, and *e*. × 1130.

B.—As in A. Septate runners. Binucleate cells usual as at *a* and *b*. The cell, *c*, contains a pair at *c*, and an extra nucleus at *d*, while the cell, *e*, is uninucleate. × 666.

C.—From 14-day infection on *Bromus marginatus*. Drained hypha with imperfect septation. × 1130.

D.—As in C. Subepidermal mycelium which will give rise to spores. Cells *a*, *b*, *c*, *d* are still multinucleate but the nuclei are often paired. × 1130.

E.—As in C. Young spore-producing mycelium beneath epidermis. Multinucleate cell at *a*. Nearly binucleate hypha at *b*. × 1130.

F.—As in C. Binucleate subepidermal hypha. × 1130.

G.—As in C. Binucleate hypha with septum forming at *a*, *b*, and completed septa at *c* and *d*. × 1130.

keeping always some distance behind the growing tips of the runners, and a wave of spore formation follows close behind. At regular intervals along the lines of runners, successive reproductive centers appear, each of which will develop into a uredinium. The long chains of uredinia so formed give to stripe rust its name. The units of this chain are small and definitely limited in growth. There are about 14 uredinia to a centimeter, on an average. Beyond the open uredinia in each chain is a series of five or six similarly spaced and progressively younger reproductive centers, and beyond these in turn are the isolated runners forming the vanguard.

Part of a longitudinal section of a leaf extending from the edge of one young reproductive center to the next is drawn in Plate 7, A. Runners (pl. 7, A, *b*, *c*, *d*) as well as the smaller feeding hyphae (pl. 7, A, *i*, *g*, *h*) are partly septate. At *a* and at *e* a few small hyphae with dense contents are massing below the upper epidermis in preparation for spore formation. Septation is a little more advanced at these centers than in the area between. This is noticeable at *f*, where the runners are most closely septate. Both runners and the smaller feeding hyphae contribute branches to the subepidermal growth.

The steps in the development of uredinium have been followed in detail. The first hyphae to reach the epidermis are still composed of multinucleate cells. In Plate 8, D, representing a triangular subepidermal air space, the hyphal cells, *b*, *c*, *d*, have four nuclei each, and the cell, *a*, has six near the tip and one farther back. This condition soon changes, and alongside of a hypha with many nuclei in a cell (pl. 8, E, *a*) another may be found (pl. 8, E, *b*) in which, although growth and nuclear division are somewhat in advance of septum formation, the binucleate tendency is evident. Soon after this, hyphae with strictly binucleate cells (pl. 8, F) are found skirting along below the epidermis.

Little has been learned as to the way in which septa form. In Plate 8, G, the irregular surface (seen as a sinuous line in edge view) extending from *a* toward *b* may be a partly formed septum. Fully formed septa are plane (pl. 8, G, *c*) or slightly arched due to turgor differences on the two sides (pl. 8, G, *d*).

The amount of subepidermal growth preceding spore formation varies somewhat, but it suffices to pry the epidermis loose from the underlying mesophyll. Short, broad cells are formed with two nuclei and dense cytoplasm. (Pl. 9, A, *a*, *b*, *c*, *d*.) These basal cells form an almost continuous subepidermal layer, first evident at the center of (and later throughout) the little reproductive area.

EXPLANATORY LEGEND FOR PLATE 9

A.—From 14-day infection on *Bromus marginatus*. Binucleate spore-producing mycelium. Basal cells at *a*, *b*, *c*, *d*. × 1130.

B.—From infection on White Federation wheat (C. I. 4981). Young spore mother cells, *a*, *b*, at the edge of a uredinium. × 1130.

C.—As in B. Developing urediniospore, *b*, with two nuclei and dense cytoplasm. Stalk cell at *a* × 1130.

D.—As in B. Growing spores, *a*, *b*, still thin walled. × 1130.

E.—As in B. Longitudinal section through center of uredinium, showing spores, *a*, on the upper surface, basal cells and runners, *b*, *c*, beneath, and the mesophyll tissue with intercellular mycelium and occasional dead cell, *d*. × 158.

F.—As in B. Longitudinal section showing the bridge of paraphyses, *a b*, extending from one uredinium to the next. × 158.

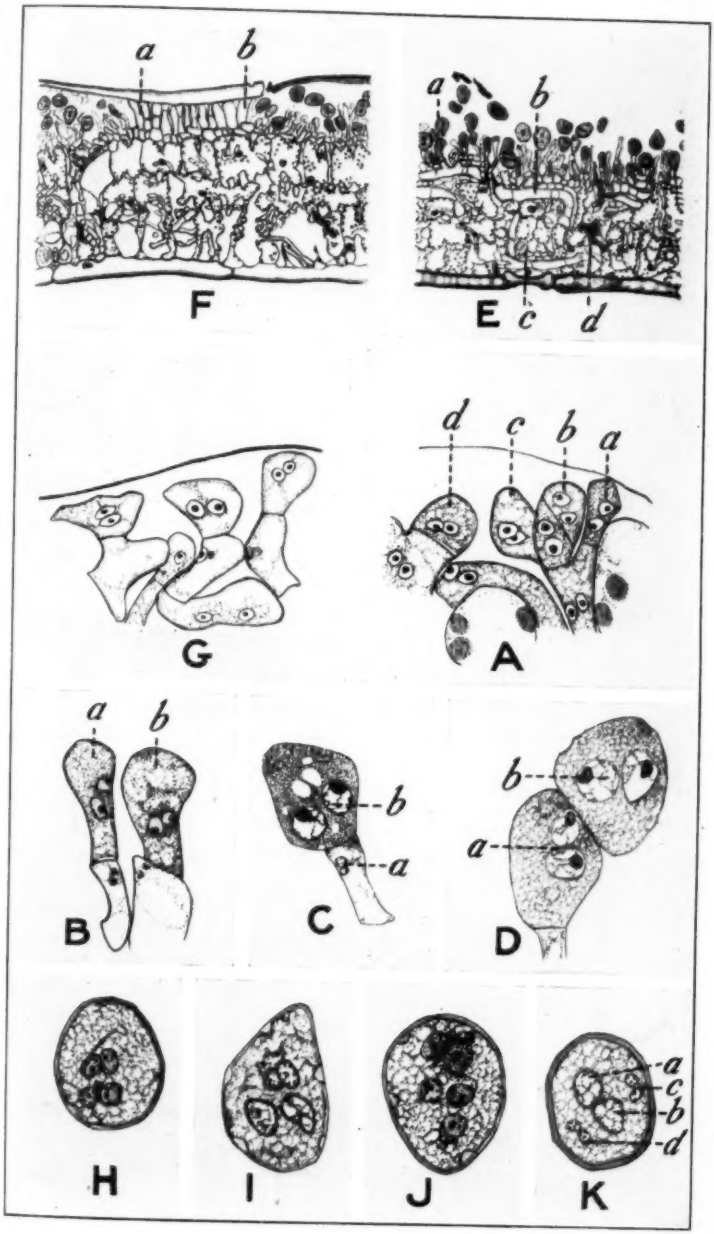
G.—From 14-day infection on *Bromus marginatus*, showing an early stage in the development of the paraphyses. × 1130.

H.—From infection on wheat C. I. 7555–1. Urediniospore with four nuclei. × 1130.

I.—As in H. Number of nuclei doubtful—probably four. × 1130.

J.—As in H. Multinucleate spore—probably six nuclei. × 1130.

K.—As in H. Spore with two nuclei, *a*, *b*, and two smaller irregular bodies, *c*, *d*. × 1130.



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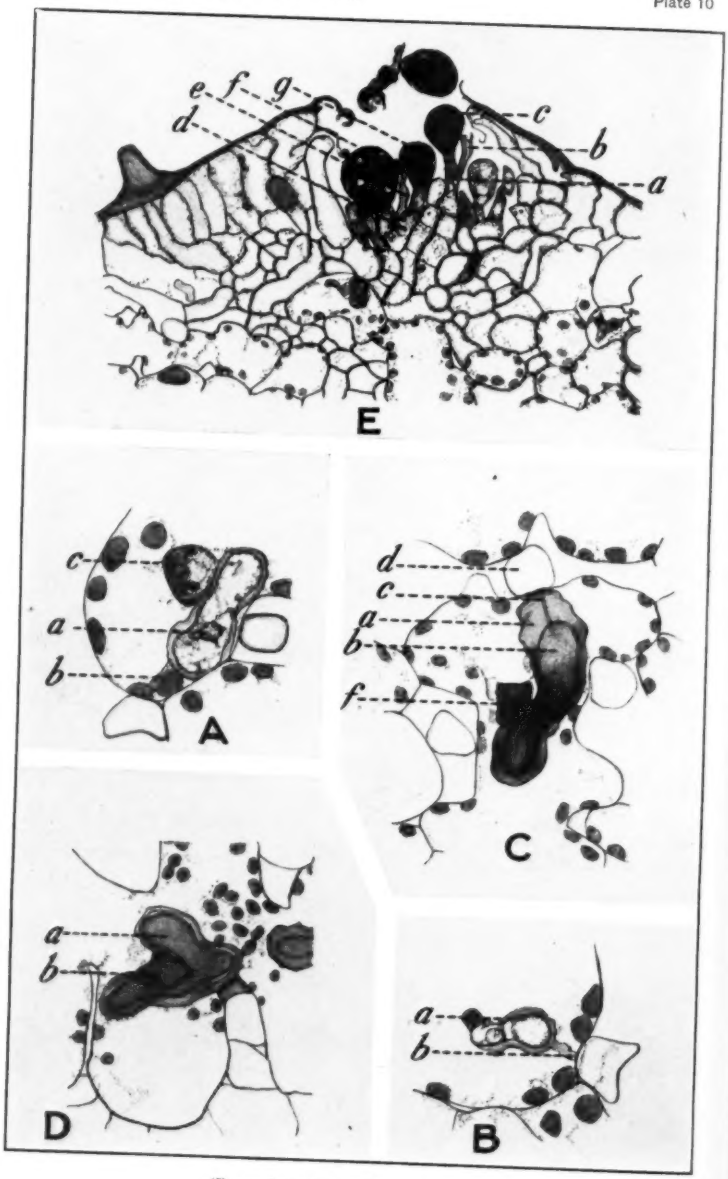

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From these the spore mother cells push up, forcing up the epidermis as they grow. (Pl. 9, B, *a*, *b*.) The spore mother cell divides into stalk cell and spore, each with two nuclei. (Pl. 9, C, *a*, *b*.) The young spore grows rapidly (pl. 9, D), and since it remains thin walled until nearly mature, the details of its contents may be seen clearly. The relative position of the two large nuclei varies. They may be in line with the longitudinal axis of the spore (pl. 9, D, *a*), at right angles to this (pl. 9, D, *b*), or in any intermediate position. The very young spores, especially if subject to lateral compression, usually have the nuclei in a longitudinal row. The mature spore, already described (pl. 1, D), usually contains dense cytoplasm and two nuclei and is covered by a wall provided with numerous scattered germ pores.

Something of the appearance of a spore-bearing region at the height of its activity may be seen in Plate 9, E, representing part of a longitudinal section through the center of an open uredinium. Runners (pl. 9, E, *a*, *b*, *c*) are still conspicuous, and smaller hyphae fill the lesser air spaces of the leaf.

The cells of the mycelium appear quite empty, the contents having drained into the spore-bearing layer. The first part of this translocation of materials may have occurred as a direct flow of cytoplasm and nuclei as such into branches growing to the surface. (Pl. 7, E, F.) Much of it, however, occurred after septation was completed and could have been achieved only by a process similar to that described in other rusts (*2*, *3*), in which the protoplasm of the hypha appears to break down into a simpler, more soluble form which can diffuse through septa. During this process the cytoplasm first decreases in quantity, and later even the nuclei become smaller and disappear, leaving the cells of the hypha free of stainable content. Haustoria as well as hyphae become drained, and in normal old age a haustorium (pl. 6, I) is nearly empty, containing but a few delicate strands of cytoplasm and the single reduced nucleus (pl. 6, I, *a*).

Not all stripe-rust haustoria undergo normal drainage. Among the haustoria beneath uredinia are some which have degenerated and died apparently without drainage. Plate 10, A and B, shows the beginnings of this change. Within each haustorium, *a*, *a*, nucleus and cytoplasm can still be seen, but each haustorium is incased in a heavy sheath, and its neck (pl. 10, A, *b*, and B, *b*) has become modified. Later (pl. 10, C) the nucleus and cytoplasm of the haustorium break down into a mass that stains heavily and uniformly. The outline of the body, *b*, of the haustorium can still be discerned within the heavy sheath, *a*, and the slender neck, *c*, can be seen leading from it to the empty haustorium mother cell, *d*, outside of the host cell. The dead host nucleus, *f*, lies in contact with the haustorium. The host cell is not collapsed nor disordered and still contains plastids

EXPLANATORY LEGEND FOR PLATE 10

A.—Early stage of degeneration of haustorium from infection in leaf sheath of heading plant of wheat, C. I. 7555–1. The haustorium, *a*, is living, but it is coated with a heavy sheath, and its neck, *b*, is modified. The host cell is living and the host nucleus, *c*, lies against the haustorium. × 1130.

B.—Similar to A. The haustorium, *a*, and its neck, *b*, are heavily sheathed. × 1130.

C.—Dead haustorium from an infection on the leaf blade of heading plants of White Federation wheat (C. I. 4981). Within the heavy sheath, *a*, can be seen the haustorium, *b*, with its neck, *c*, leading to the haustorium mother cell, *d*. The dead host nucleus, *f*, lies against the haustorium. × 1130.

D.—Similar to C. The haustorium, *a*, is branched and the host nucleus, *b*, lies at the forking. × 1130.

E.—Portion of cross section of glume of wheat, C. I. 7555–1, showing small uredinium. The basal cell, *a*, gave rise to two spores, *b* (freed) and *c*. The basal cell, *d*, formed three spores, *e* (set free), *f*, and *g*. × 510.

and cytoplasm. Plate 10, D, *a*, shows a degenerate branched haustorium with the host nucleus, *b*, between the branches. Plate 11, A, is still further degenerated and not even the neck is distinguishable. These degenerate haustoria occur in old infections on all hosts studied, even though the rust has made good growth. In one advanced infection on old leaf-sheath tissue of wheat (C. I. No. 7555–1) 50 per cent of the haustoria showed this type of breakdown.

The spores are borne chiefly on the upper surface (pl. 9, E) and are at all stages of development. Just beneath them are the short broad basal cells from which they arise. The first-formed spores have been freed, leaving the withered spore stalks behind them. The younger spores must force their way up between the stalks of older spores, often under considerable pressure.

The same basal cell can give rise to several spores in succession. This shows clearly in Plate 10, E, a cross section through a small uredinium growing on a glume of wheat. The basal cell, *a*, first formed a spore at *b* (of which only the withered stalk is left) and then a second, *c*. The basal cell, *d*, formed three spores, one at *e* (evidenced by the stalk), a second at *f*, now mature, and a third, at *g*.

About the margin of the uredinium is more or less of sterile fungous tissue composed of coarse upright cells set side by side in the fashion of a palisade. These paraphyses vary greatly in number. In some infections, particularly on seedling hosts, they are practically absent. In others, each uredinium is completely girt about with sterile tissue. There is an abundance of it on either side of the small uredinium in Plate 10, E. In some chains of uredinia on nearly mature host plants there is a bridge of sterile tissue between each uredinium and the next. (Pl. 9, F, *a* to *b*.) These upright sterile cells originate in a line of basal cells similar to those at the base of the spores. When very young, this marginal sterile tissue (pl. 9, G) resembles closely in structure the young spore-bearing tissue adjoining it. The sterile tissue can be distinguished at a very early stage, however, by the vacuolate cytoplasm of its cells. The watery contents of these cells contrast sharply with the dense contents of fertile tissue at a corresponding age. (Cf. pl. 9, A and G.)

In one lot of material (nearly mature wheat plants fixed at Davis, Calif., in May, 1926) a small percentage of the last generation of urediniospores—those contemporaneous with the teliospores—contain more than two nuclei. Plate 9, H and I, shows spores with four nuclei. In Plate 9, J, the number is doubtful, but it may be more than four. In some cases two large central nuclei are found (pl. 9, K, *a* and *b*) and in addition smaller irregular bodies (K, *c*, *d*) similarly stained. These are unexplained. The percentage of these multinucleate spores is always small. When, during the following year

EXPLANATORY LEGEND FOR PLATE 11

A.—From 17-day infection on Jenkin club wheat. Dead haustorium, *a*. × 1130.

B.—From infection in leaf sheath of *T. vulgare* C. I. 7555–1. Green intracellular body, *a*, in living cell of host. Haustorium at *b*. × 1130.

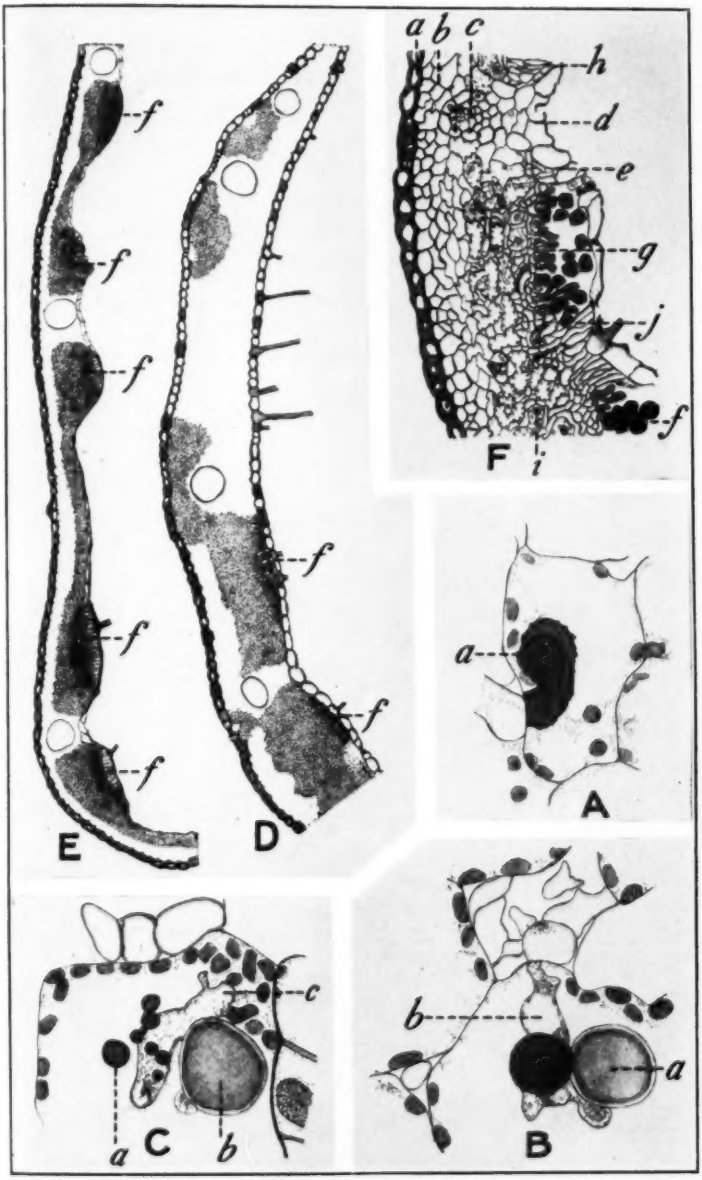
C.—From infection in leaf sheath of *T. vulgare* C. I. 7555–1. Two types of intracellular bodies—at *b* a large green body and at *a* a group of small spherical red-stained bodies. × 1130.

D.—Cross section of upper end of glume of C. I. 7555–1. Semidiagrammatic. × 79.

E.—Cross section through lower part of same glume. × 79.

In D and E, the circles represent veins, the dotted area mesophyll, and the clear area represents rather heavy-walled empty cells without intercellular spaces. Rust occurs at points marked *f*.

F.—Portion of cross section of glume of C. I. 7555–1 showing outer epidermis at *a*, lined by strengthening tissue, *b*, which extends around vein, *c*, to inner epidermis at *d*. Mesophyll tissue containing uredinia at *f*, *g*, and *i*, flanked by paraphyses at *e* and *j*. Edge of next patch of mesophyll and fungus at *h*. × 158.



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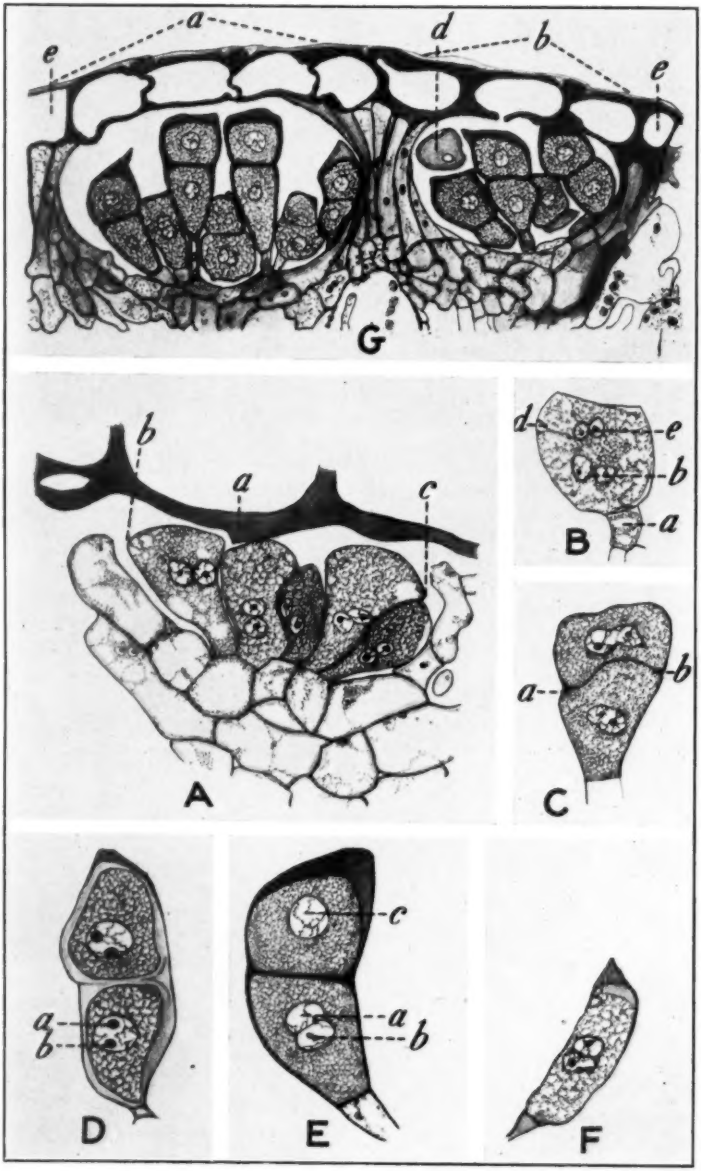

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(June, 1927), telia of stripe rust appeared on plants of *Bromus marginatus* in Berkeley, Calif., material of telia and adjoining uredinia was fixed. Urediniospores here, so far as observed, are uniformly binucleate.

The host tissues underlying uredinia (pl. 9, E, F) are generally living but impoverished. There is less evidence of the stimulation of invaded cells than in the other wheat rusts studied (*2*, *3*). Host nuclei expand but little and die prematurely. In one infection the larger nuclei in cells containing haustoria averaged 10μ by 7.5μ as compared with 8.5μ by 7.7μ in the adjoining uninfected tissue. Plastids decrease in size, and in no case has excess starch been noted. Occasionally a mesophyll cell collapses into a little irregular red-staining mass. (Pl. 9, E, *d*.) These dead collapsed cells increase in number with the age of the uredinium. There may be 10 or 15 per cent of them beneath older uredinia.

In one lot of material taken from leaf sheaths of nearly mature wheat (C. I. No. 7555–1), two types of intracellular bodies were found. One (pl. 11, B, *a*) is spherical, ranges in size up to 12μ, stains green in the triple stain, is nearly transparent, and is homogeneous in content save for a paler outer layer. It may have any position in the cell. These green-staining spheres are abundant in the infected tissue but also have been seen in a few cells just beyond the margin of the mycelium. Since tissue more remote from the infection was not fixed, it is not certain that these bodies are due to the activities of the rust.

The other type of intracellular body is small, spherical, and stains a deep red. In Plate 11, C, both types are to be found in the same cell, the green-stained body at *b* and a group of little red-stained spheres at *a*. Both types are also to be seen in Plate 6, H, already referred to. These red globules have, so far, not been seen outside of infections. They are most numerous in cells with little protoplasm, and their formation is apparently associated with the breakdown of living matter under the influence of the fungus.

Puccinia glumarum, as already mentioned, can grow and reproduce in the glumes. Uredinia occur in rows running parallel to the veins of glumes and even of awns. In the glumes, nearly all of these uredinia open upon the inner face, so that the spores are shed into the narrow crevice between the glume and the inner flower parts, often lodging and accumulating there. This is a disadvantage to the rust, as such spores are not readily caught and dispersed by the wind.

The distribution of stripe rust in glumes is limited by three facts: The fungus can enter only through stomata; it can spread only through intercellular spaces; and it can form haustoria only in relatively thin-walled living mesophyll tissue and epidermis. In the

EXPLANATORY LEGEND FOR PLATE 12

A.—Part of a section of a leaf sheath of wheat C. I. 7555–1, representing a young telium, with spore initials, *b c*, with large nuclei and dense cytoplasm. Below are the basal cells, at the sides are young paraphyses and above is the epidermis, *a*. × 1130.

B.—A young teliospore (on C. I. 7555–1) with stalk cell, *a*, and distal cell containing two pairs of nuclei, *b c*. A septum which will divide it into two cells is forming at *d*. × 1130.

C.—Growing teliospore from C. I. 7555–1. Nuclear fusion is in progress. The wall is still thin except at ends of septum, *a b*. × 1130.

D.—Nearly mature teliospore (on C. I. 7555–1) with dense cytoplasm and heavy walls. The fusion nucleus of each cell still contains two nucleoles, *a*, *b*. × 1130.

E.—Mature teliospore (on C. I. 7555–1) showing delayed nuclear fusion in one cell, *a b*, and completed fusion in the other, *c*. × 1130.

F.—Single-celled teliospore (on C. I. 7555–1) at edge of sorus. × 1130.

G.—Section (of C. I. 7555–1) through two mature telial units, *a*, *b*, each containing a compact group of spores springing from a pseudoparenchymatous base of fungous cells, inclosed laterally by upright, somewhat compressed paraphyses, and roofed by the epidermis of the host, *e e*. Urediniospore at *d*.

wheat glumes examined (C. I. No. 7555–1) much of the interior is made up of strengthening tissue composed of empty, rather thick-walled cells without intercellular spaces. The fungus can not penetrate this tissue.

Plate 11, D, represents semidiagrammatically part of a cross section through the upper end of a glume and Plate 11, E, one from the lower third of a glume. In both, the circles represent veins, the dotted areas mesophyll, and the clear areas correspond to the strengthening or mechanical tissue. In the lower part of the glume (pl. 11, E) the strengthening tissue surrounds the veins and forms a continuous sheet lining the outer (convex) epidermis. The mesophyll faces the inner epidermis between veins and is thickest near the veins. In the upper part of the glumes the arrangement is more irregular. Part of the mesophyll fronts outward. The outer epidermis has thick walls throughout. The inner epidermis is thinner walled—markedly so in the lower, less-exposed part. So far as noted, stomata occur only where the epidermis is in contact with mesophyll.

So the rust (points marked *f* in the diagrams) enters only where there is mesophyll, spreads only through its spaces, and grows out to the inner surface (the only one accessible except at the tip of the glume) to form uredinia.

The tissues of the glume are drawn at higher magnification in Plate 11, F. At *a* is the heavy-walled outer epidermis, and lining it is a sheet of strengthening tissue, four or five cells thick, free of fungus. This tissue extends around the vein, *c*, and on to the inner epidermis at *d*. Under the inner epidermis from *e* to *f* is mesophyll permeated by fungus. There is one small uredinium at *g* and the edge of a second at *f*. There is a bridge of paraphyses between the two. At *h*, above the vein, are paraphyses adjoining a third uredinium. Plate 10, E, already described, is also from a glume.

DEVELOPMENT OF TELIA

Telia are formed on host plants nearing maturity. They occur on the leaf sheaths, on the glumes, and, more rarely, on the leaf blades. A brief study has been made of telia on the leaf sheath, where they are most abundant. The small compact telia form along the edges of old uredinia. Exceptionally, the two kinds of spores may be found mixed in the same sorus.

A typical young telium is drawn in Plate 12, A. The cells that are to become spores, *b* to *c*, are easily distinguished by their dense contents and large nuclei. Their form varies with the available space. They may be misshapen by pressure against the heavy epidermis. When relatively free, as in this case, they are balloon shaped.

In development, the initial cell first divides, forming a short stalk cell. (Pl. 12, B, *a*.) Then the two nuclei in the spore proper undergo division, forming four nuclei. (Pl. 12, B, *b*, *c*.) In this case the septum which will divide the spore into two cells is beginning to form at *d*.

As the spore grows and takes shape, the two nuclei in each cell fuse. This process may begin as soon as the spore is two celled and in some cases is not complete until the spore is nearly mature. In Plate 12, C, a half-grown spore, still thin walled, shows an early stage of fusion. In each cell the two nuclei have come in contact, and the nuclear membrane at the surface of contact has disappeared. The two sets of chromatin still appear to be distinct. Later the fusion nucleus

becomes round but may still show the nature of its origin by the presence of the two nucleoles. (Pl. 12, D, *a*, *b*.)

Slight irregularities occur. Sometimes fusion is delayed in one cell (pl. 12, E, *a*, *b*), although completed in the other (pl. 12, E, *c*). Occasionally one finds a one-celled teliospore (pl. 12, F) and more rarely a three-celled one.

Something of the appearance of the mature telia is shown in Plate 12, G. Each of the two compact telial units, *b* and *a*, is roofed by the heavy epidermis of the host, *e*, and walled in at the sides by a palisade of paraphyses similar to that surrounding the uredinia. The spores are packed closely together and shaped to some extent by the available space. At *d* is part of a urediniospore. Farther on in this sorus urediniospores predominate.

DISCUSSION

The earliest characteristic difference found between stripe rust and other wheat rusts is the absence of a well-marked appressorium. This was also noted by Pole Evans (*21*). The contents of the germ tube are somewhat denser at the growing tip, but the tube swells little, if at all, on reaching the stoma, and ordinarily there is no septum marking off a separate appressorial cell. Even before entering the host the tendency in stripe rust toward the minimum of cross walls finds expression. This procedure would appear to be a disadvantage to the rust, for in cases of delayed entry a thin-walled germ tube exposed to wind and sun would probably stand a poorer chance of survival than the compact appressorial cushion of other rusts.

The incubation period in stripe rust of wheat is from two to four days longer than in either of the other wheat rusts. Urediniospores germinate overnight, yet inoculated leaves do not "fleck" until the eighth, ninth, or even tenth day (varying somewhat with the host and the time of year). A part of this delay is due to the prolonged juvenile period of the rust. On the second day the fungus consists of a small substomatal vesicle and one to three short slender infecting hyphae, each of which has formed a little haustorium. During the next two days the only development is a swelling of the vesicle and initial hyphae to several times their original dimensions. Since these are only 1 or 2 or, at most, 3 little haustoria to draw food from the host, this process is necessarily slow.

Rice (*35*) finds in early infection stages of corn rust a well-developed intercellular mycelium before the development of haustoria, and concludes that the fungus is not entirely dependent on the haustorium for its nutrition. In stripe rust, on the contrary, the conspicuous bulging of the first infecting hyphae just back of the haustorium mother cells gives ample evidence that the food is coming in through these mother cells from the haustoria.

No evidence of direct feeding by the hyphae has been noted in wheat rusts. Haustoria are formed at the beginning of infection and increase in number in direct proportion to the development of the mycelium. They seem adequate for the task of absorbing food for the parasite, but, of course, it would be difficult either to prove or disprove that a small amount of food was absorbed directly by the hyphae. If, in some rusts, a part of the food is absorbed directly by the hyphae without the formation of the special absorbing organs,

the haustoria, the hitherto baffling problem of growing rusts in culture media looks a little less impossible of solution.

It is of interest that the exceptional size of stripe-rust hyphae is not attained gradually as the fungus grows out into the tissue and establishes more contacts with the host, but is initiated right at the outset of the vegetative life. The first hypha pushed out from the expanded fungous mass at the stoma of entry is full sized and is multinucleate and nonseptate like the later hyphae.

Rice (*35*) and the writer (*3*) compiled from rust literature data on the number of nuclei in haustoria of gametophytic and sporophytic mycelia. The numbers as reported are somewhat variable, but uninucleate haustoria predominate in the gametophyte and binucleate in the sporophyte. Wheat rusts do not conform to this. *Puccinia graminis* (*2*), *P. triticina* (*3*), and *P. glumarum* have regularly uninucleate haustoria in the uredinial (sporophytic) generation. In *P. glumarum* only two haustoria have been seen which had two nuclei, and none with more. The haustorium mother cells have a variable nuclear content, but apparently all but one of these nuclei degenerate before the haustorium is formed.

The restrictions in the spread of stripe-rust mycelia imposed by host tissues impenetrable to the fungus were fully described by Eriksson and Henning in "Getreideroste" (*20*). Hursh (*26*), in his study of the relation of morphologic structure of the wheat plant to resistance to *Puccinia graminis tritici*, found a similar effect on the progress and ultimate development of the invading fungus. These limitations, together with the production by stripe rust of vigorous runners which find their freest path in the open spaces beneath the rows of stomata, determine the course of progress and the final form of the infection.

The active period of any one portion of an infection is brief. By the time the host tissues beneath one uredinium are impoverished, new uredinia down the line are shedding spores. The impoverishment of infected host tissues takes place somewhat faster in stripe rust than in stem and leaf rusts of wheat. Moreover, degenerate haustoria, hitherto regarded (*2*) as an indication of uncongenial relations between host and rust, are found in older parts of infections on all the hosts studied.

Perhaps the more rapid breakdown of relations between host and pathogene in any given area is to be correlated with the unusual ability of the latter to move on to fresh areas. At any rate, this continued migration gives added efficiency and increased spore production.

In stripe rust there is associated with an ability to form an indefinite series of uredinia a definite limitation in the size of the individual uredinium. The uredinia of many rusts spread marginally, forming new spore-bearing cells around the edges as the central part nears exhaustion. In stripe rust the definite little spore-bearing area usually is bordered by sterile cells which are probably homologous to spores (or spore mother cells when the final septum is omitted) and whose appearance suggests a half-eradicated tendency toward the marginal growth of the uredinium common in many rusts. The function of these paraphyses is doubtful. They may perhaps be of some service as buffer tissue, helping to raise the epidermis and to release the young spores from its pressure. It should be mentioned, however,

that uredinia lacking this sterile tissue—and such are not infrequent—show no lack of effectiveness.

On one of the hosts studied, intracellular bodies of two types are found in infected areas. Beauverie (*8*) describes several types of intracellular bodies found in degenerating plant tissues, some of which are formed in cells containing fungous haustoria. He suggests that the "nucleoles" which constituted for Eriksson the histologic proof of mycoplasm are nothing but intracellular bodies of one or more types.

Eriksson's figures (*17*) show some of the "Plasmanucleoli" (supposed mycoplasm) as little red-stained globules lying free in the cell and others as connected with the fungus outside by a fine tube extending to and through the host cell wall. The free globules were probably red-stained intracellular bodies of the sort figured in this paper (pl. 11, C), and the others were undoubtedly early stages in the formation of haustoria. The spherical heads of very young haustoria stain deep red with the triple stain.

Another type of intracellular body was figured by Ruttle and Fraser (*36*) in infections of *Puccinia coronata* Cda., and judging by the figures, this same type was seen by Zach (*39*) in infections of *P. glumarum*.

Prior to septation, the nuclei are irregularly disposed, isolated or in groups, along the length of the hyphae. When the time for septation comes, these nuclei sort themselves into pairs which become fairly uniformly distributed. The basis of this assortment is unknown, but it is possible that there is here a pairing of "plus" and "minus" elements such as is known in Phycomycetes, Ascomycetes, Ustilaginales, and Agaricales, and is now announced by Craigie (*13*) in one of the rusts; and that somehow the pairs of nuclei in stripe rust are comparable to the pairs in binucleate mycelium of other rusts. When one considers, however, that no aecial stage, and hence no fusion of uninucleate cells, is known in stripe rust, the mechanism of this process becomes obscure. Of course, even if (as is not proved) stripe rust lost its aecial stage long ago, whatever basis of assortment originally obtained might still persist.

When the binucleate condition is once established, further development parallels that of other rusts. The new cells formed agree closely in size, form, and activities with the corresponding stages in rusts with regularly binucleate mycelium, and binucleate spores are formed in the usual fashion.

In uredinia adjoining telia in one lot of material, a few of the urediniospores contain more than two nuclei. It is doubtful what weight to give this. These multinucleate spores have been seen so far only in association with telia, and this aroused a suspicion that stripe rust, whose aecial host is yet to be found, may be executing some short cut in its life history by which a small percentage of spores in the final uredinial generation on a maturing host function as teliospores. On this basis, the young urediniospore should contain two nuclei, then one, then two again, and finally four. Evidence on this point was not available, for the uredinia adjoining the telia in this material were old, and very few young urediniospores were present. During the next year, when an attempt was made to secure a slightly earlier stage, only binucleate urediniospores were found.

On the other hand, stripe rust has multinucleate cells through much of its history, and the presence of more than two nuclei in a urediniospore may be only one more expression of this mode of life. It is consistent with the rest of the life history.

The brief study of the development of the teliospore included here suffices to show that it is quite regular and like that of other rusts. The germination of stripe-rust teliospores and the formation of sporidia have been described by Eriksson and Henning (*20*). No deterioration in this part of the cycle has been observed, yet, so far as known, it is useless. The fact that teliospores and sporidia continue to be formed year after year can not be taken as proof of the existence of an aecium. Elsewhere, functionless structures have persisted, as, for example, in regularly apogamous ferns (*1*) which continue to form and liberate active spermatozoids. In other words, defective telia and sporidia would have constituted evidence against the presence of an aecium in the life cycle, but normal telia and sporidia do not prove its existence.

No process has been seen in the uredinial generation of stripe rust which would compensate for a missing aecial stage. Of course, among the hundreds of nuclei in the large vegetative hyphae, nuclear fusion and reduction could take place undetected, but its occurrence at that stage is improbable. With the doubtful exception of four-nucleate urediniospores, no positive evidence has been noted which would lead to a belief in the absence of an aecial stage.

SUMMARY

Urediniospores of *Puccinia glumarum* contain two, or rarely more, nuclei. The spore wall has 10 to 16 germ pores. On germinating, the germ tube grows to the stoma and enters without forming a definite appressorium.

The substomatal vesicle is multinucleate. It pushes out one to three short infecting hyphae, each of which forms a small haustorium. On the second, third, and fourth days the vesicle and infecting hyphae expand to several times their original volume.

From this expanded mass at the stoma of entry, a hypha grows out into the intercellular spaces of the leaf tissue. Young hyphae are coarse, multinucleate, and, apart from haustorium formation, nonseptate.

Haustoria are formed in abundance. The haustorium mother cell is a short, broad, terminal cell of a hypha, containing two to six nuclei. The haustorium is uninucleate, simple or branched, and has a maximum length of 35μ or 40μ. The host nucleus is usually associated with the haustorium. In normal old age the haustorium becomes drained and nearly empty. Some of the haustoria of older infections on all hosts studied undergo degeneration. The necks of others become encrusted with granular, dark-stained matter.

Vigorous nonseptate runners grow lengthwise of the leaf through substomatal passageways, spreading the rust. Branches from these runners permeate the smaller air spaces of the tissues. The transverse spread is limited by leaf veins reinforced by strengthening tissue impenetrable to the fungus.

About the tenth day after inoculation, septation begins in the older parts of the mycelium, and thereafter the septate area spreads, keeping a short distance behind the advancing runners. The first

septa cut a hypha into long cells, then nuclei (hitherto irregularly disposed) become arranged in pairs distributed along its length, and further septa cut it into binucleate cells.

During septation, branches from both the runners and the feeding hyphae grow to the upper epidermis and form there subepidermal binucleate spore-bearing cells. Urediniospores are usually binucleate. A basal cell may give rise to several spores in succession. A few of the last urediniospores formed on a maturing host plant may be multinucleate. During spore formation the host tissues beneath become impoverished and the mycelium drained. Intracellular bodies of two types are found in infections on one host.

Uredinia are small and definitely limited in size and are usually bounded by a zone of paraphyses. This limitation in the size of the individual uredinium is compensated by the production of many uredinia in succession along the lines of advance of the runner. Chains of one hundred or more uredinia are not rare.

In glumes, much of the tissue is impenetrable to the hyphae. The mesophyll tissue usually faces the inner surface of the glume and the stomata occur in epidermis adjoining mesophyll tissues, so the rust usually enters from the inner surface, spreads through the mesophyll, and comes to the inner surface to form uredinia.

Telia may form as the host matures. The development of the teliospore is normal. The spore initial divides, forming a stalk cell, and divides again, forming a two-celled spore. Each cell contains two nuclei which fuse as the spore matures.

No positive evidence has been noted of any compensatory process in the uredinial generation which would replace an aecial stage.

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THE GROWTH OF RHODE ISLAND REDS AND THE EFFECT OF FEEDING SKIM MILK ON THE CONSTANTS OF THEIR GROWTH CURVES[1]

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INTRODUCTION

The purpose of the study here reported was to obtain data regarding the growth of Rhode Island Red cockerels, capons, and pullets, and to determine what effect the feeding of skim milk would have on their growth. Inasmuch as other investigators who have made similar studies have said little or nothing about the variability of their data, it was decided that the present study should include this feature.

In order to obtain the desired data it was proposed that two similar groups of chicks be used and that one group be fed a ration presumably adequate in every way for normal growth and that the other group be fed the same ration and in addition be allowed free access to skim milk at all times.

EXPERIMENTAL PROCEDURE

Two groups of chicks were used—one of 174, which was hatched March 14, 1924, and another of 191, hatched 10 days later. Each chick in each group was weighed as soon after hatching as possible and at age intervals of one week until 18 weeks old and thereafter every other week. The feed placed before the two groups was identical from the start, except that the group hatched last had access to sour skim milk and water at all times, whereas the one hatched first was permitted to drink only water.

The first group was put into a colony brooder house, 10 by 10 feet, and allowed access to a grass yard about 60 by 75 feet. The chicks were thus reared until they were 10 weeks old, at which age the sexes were separated. Until that time the mortality was 20 chicks. Of those living, 80 were pullets and 74 were cockerels. The latter were divided as evenly as possible in respect to weight, and 40 were caponized, 3 dying following the operation. Fifty-five pullets were selected by taking every other one as they were caught, the remainder being discarded in order to reduce the pullets to more nearly the same number as the cockerels and the capons. Thus at the beginning of the eleventh week there were 34 cockerels, 37 capons, and 55 pullets. Each lot, now designated as lot 1 cockerels, lot 1 capons, and lot 1 pullets, was put in a colony brooder house, 10 by 10 feet, surrounded by a grass yard about 60 by 35 feet. The mortality

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from the beginning of the eleventh week to the end of the period the chicks were under observation was as follows:

Lot 1 cockerels (eleventh to thirty-fourth week)	3
Lot 1 capons (eleventh to thirty-eighth week)	2
Lot 1 pullets (eleventh to twenty-sixth week)	2

Thus the total number in each of the lots on which growth observations were made from hatching time to the end of the experiment was: 31 cockerels, 35 capons, and 53 pullets.

The second group received the same treatment as the first group. At the end of the tenth week the mortality was 28 chicks. Of the surviving chicks 80 were pullets and 83 were cockerels. The latter, as before, were divided into two groups and 39 were caponized, two dying following the operation. Forty pullets were again selected in the manner described. There were, then, at the beginning of the eleventh week: 44 cockerels, 37 capons, and 40 pullets. These lots, from this point on, were designated as lot 2 cockerels, lot 2 capons, and lot 2 pullets. For the remainder of the period that the chicks were under observation the mortality was as follows:

Lot 2 cockerels (eleventh to thirty-fourth week)	4
Lot 2 capons (eleventh to thirty-eighth week)	3
Lot 2 pullets (eleventh to twenty-sixth week)	2

Thus the total number in each of the lots on which growth observations were made from hatching time to the end of the experiment was: 40 cockerels, 34 capons, and 38 pullets.

All six lots of chicks received the same treatment and the same feed throughout, with the one exception that in addition to the solid feed fed lot 2 cockerels, lot 2 capons, and lot 2 pullets had free access at all times to sour skim milk and water, whereas lot 1 cockerels, lot 1 capons, and lot 1 pullets were allowed only water. For the first two weeks a slightly moistened mash was fed three times daily; this mash consisted of:

	Parts by weight
Corn meal	2
Oats	2
Middlings	1
Bran	1
Whole boiled egg (including shell)	1

A commercial chick scratch feed was also fed twice daily.

After the first two weeks a dry mash was kept before the chicks in self-feeding hoppers; this mash was mixed as follows:

	Parts by weight
Corn meal	3
Crushed oats	2
Middlings	1
Bran	1
Meat scrap (60 per cent)	1

In addition to the above mash the chick scratch feed was fed three times daily until the chicks were 4 weeks old; at that age a change was made to a scratch feed containing equal parts of coarsely cracked corn and wheat. Each lot, as previously noted, had access to grass range.

THE GROWTH DATA

Each chick, as before stated, was weighed every week until 18 weeks old and thereafter every other week. In order to determine the vari-

ability of the live weights of the chicks in each lot, the average live weight, the standard deviation, and the coefficient of variation were calculated. For the purpose of facilitating comparison of the average growth of each lot, the average live weights are brought together and presented in Table 1. In Table 2 the probable error of the mean (average) live weight, the standard deviation and its probable error, and the coefficient of variation and its probable error are given.

TABLE 1.—*Average weight of the chicks in each lot at the ages indicated*

Age (weeks)	Average weight of—					
	Cockerels		Capons [a]		Pullets	
	Lot 1	Lot 2	Lot 1	Lot 2	Lot 1	Lot 2
	Grams	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*
0	40.40	39.82	40.86	40.73	39.69	38.99
1	63.95	62.99	62.88	65.85	61.87	62.64
2	85.78	99.22	83.88	98.26	81.37	94.00
3	110.27	163.33	106.57	161.84	104.02	148.60
4	131.84	218.33	132.27	219.80	121.89	194.06
5	173.96	317.62	173.77	312.79	158.04	281.58
6	245.81	449.50	242.50	442.35	212.45	396.71
7	317.42	542.37	304.43	530.23	269.72	471.05
8	422.52	665.50	401.43	657.35	352.32	578.76
9	521.45	815.37	498.51	808.97	437.83	695.00
10	599.84	973.37	579.77	981.03	505.98	817.50
11	713.39	1,154.75	695.86	1,128.09	594.81	942.10
12	879.84	1,321.12	813.14	1,278.09	725.47	1,049.34
13	1,052.90	1,474.32	979.14	1,456.47	844.53	1,134.87
14	1,234.84	1,628.50	1,104.14	1,607.09	966.70	1,261.71
15	1,393.39	1,744.12	1,266.00	1,714.85	1,075.94	1,340.53
16	1,497.58	1,834.37	1,386.57	1,876.32	1,176.98	1,427.89
17	1,630.80	1,961.81	1,532.07	2,037.79	1,276.32	1,536.21
18	1,764.03	2,089.25	1,677.57	2,199.26	1,375.66	1,644.74
20	2,113.55	2,300.12	2,034.28	2,367.94	1,591.89	1,747.24
22	2,274.19	2,472.25	2,313.28	2,595.59	1,692.45	1,919.34
24	2,559.84	2,650.00	2,539.43	2,782.79	1,880.00	2,074.60
26	2,706.61	2,799.62	2,660.28	2,828.38	2,009.81	2,137.37
28	2,931.93	2,920.50	2,893.43	2,955.44	----------	----------
30	3,050.81	3,081.37	2,946.43	3,118.32	----------	----------
32	3,254.03	3,188.75	3,072.28	3,257.65	----------	----------
34	3,322.58	3,437.00	3,171.00	3,346.47	----------	----------
36	----------	----------	3,320.71	3,403.97	----------	----------
38	----------	----------	3,457.14	3,556.03	----------	----------

[a] Caponized when 10 weeks old.

TABLE 2.—*Statistical constants of the data given in Table 1*

Age (weeks)	Statistical constants	Cockerels		Capons		Pullets	
		Lot 1	Lot 2	Lot 1	Lot 2	Lot 1	Lot 2
0	P. E. of mean..gm..	± 0.46	± 0.33	± 0.48	± 0.51	± 0.33	± 0.44
	σ.....gm..	3.83± .33	3.15± .24	4.23± .34	4.41± .36	3.52± .23	4.04± .31
	C. of V..per cent..	9.48± .81	7.91± .60	10.35± .84	10.83± .90	8.87± .58	10.36± .81
1	P. E. of mean..gm..	± 1.18	± 1.19	± .87	± 1.26	± .87	± 1.16
	σ.....gm..	9.58± .83	11.17± .84	7.56± .62	10.93± .89	9.43± .62	10.64± .82
	C. of V..per cent..	14.98± 1.33	17.73± 1.38	12.02± 1.00	16.60± 1.40	15.24± 1.02	16.98± 1.35
2	P. E. of mean..gm..	± 2.30	± 2.14	± 1.13	± 2.08	± 1.25	± 1.99
	σ.....gm..	19.01± 1.63	20.05± 1.51	9.96± .80	18.01± 1.47	13.51± .88	18.15± 1.40
	C. of V..per cent..	22.16± 1.99	20.21± 1.59	11.94± .98	18.33± 1.55	16.60± 1.12	19.31± 1.55
3	P. E. of mean..gm..	± 3.15	± 3.64	± 2.12	± 3.65	± 1.80	± 3.55
	σ.....gm..	25.99± 2.23	34.13± 2.57	18.32± 1.50	31.54± 2.58	19.09± 1.27	32.45± 2.51
	C. of V..per cent..	23.57± 2.13	20.90± 1.64	17.19± 1.45	19.49± 1.65	18.35± 1.27	21.84± 1.77
4	P. E. of mean..gm..	± 3.92	± 4.59	± 2.82	± 5.24	± 2.16	± 5.01
	σ.....gm..	32.39± 2.77	43.01± 3.24	24.78± 2.00	45.34± 3.71	23.33± 1.53	45.81± 3.54
	C. of V..per cent..	24.57± 2.23	19.70± 1.54	18.73± 1.56	20.63± 1.76	19.14± 1.30	23.61± 1.93

TABLE 2.—*Statistical constants of the data given in Table 1*—Continued

Age (weeks)	Statistical constants	Cockerels		Capons		Pullets	
		Lot 1	Lot 2	Lot 1	Lot 2	Lot 1	Lot 2
5	P. E. of mean gm.	± 5.45	± 6.24	± 3.24	± 6.98	± 3.01	± 6.00
	σ per cent.	44.97± 3.85	58.49± 4.41	28.44± 2.29	60.31± 4.93	32.46± 2.13	54.86± 4.24
	C. of V. per cent.	25.85± 2.36	18.41± 1.44	16.37± 1.36	19.28± 1.64	20.54± 1.40	19.48± 1.56
6	P. E. of mean gm.	± 9.41	± 8.60	± 5.40	± 8.97	± 4.75	± 8.33
	σ gm.	77.69± 6.65	80.67± 6.08	46.67± 3.82	77.57± 6.34	51.27± 3.36	76.16± 5.89
	C. of V. per cent.	31.60± 2.97	17.95± 1.40	19.24± 1.63	17.53± 1.48	24.13± 1.67	19.20± 1.54
7	P. E. of mean gm.	±13.01	± 8.93	± 6.67	± 8.21	± 5.99	± 7.95
	σ gm.	107.40± 9.20	83.75± 6.31	58.50± 4.72	70.99± 5.81	64.66± 4.24	72.70± 5.62
	C. of V. per cent.	33.83± 3.21	15.44± 1.19	19.22± 1.62	13.39± 1.12	23.97± 1.66	15.43± 1.22
8	P. E. of mean gm.	±15.70	±10.35	± 8.69	± 9.27	± 8.58	± 9.43
	σ gm.	129.58±11.10	97.06± 7.32	76.21± 6.14	80.10± 6.55	91.79± 6.01	86.21± 6.67
	C. of V. per cent.	30.67± 2.86	14.58± 1.12	18.98± 1.58	12.18± 1.01	26.05± 1.82	14.89± 1.18
9	P. E. of mean gm.	±17.81	±12.35	±10.38	±10.12	±10.21	±10.43
	σ gm.	147.03±12.59	115.77± 8.73	91.04± 7.34	87.48± 7.15	110.22± 7.22	95.37± 7.38
	C. of V. per cent.	28.20± 2.60	14.20± 1.09	18.26± 1.52	10.81± .90	25.17± 1.75	13.72± 1.08
10	P. E. of mean gm.	±19.45	±13.62	±11.11	±12.66	±11.27	±12.42
	σ gm.	160.55±13.75	127.72± 9.63	97.48± 7.86	109.43± 8.95	121.69± 7.97	113.47± 8.78
	C. of V. per cent.	25.76± 2.45	13.12± 1.01	16.81± 1.39	11.15± .92	24.05± 1.66	13.88± 1.09
11	P. E. of mean gm.	±21.39	±15.85	±12.98	±14.50	±12.84	±13.77
	σ gm.	176.56±15.12	148.58±11.20	113.82± 9.18	125.33±10.25	138.58± 9.08	125.83± 9.74
	C. of V. per cent.	24.75± 2.25	12.87± .99	16.36± 1.35	11.11± .92	23.30± 1.61	13.36± 1.05
12	P. E. of mean gm.	±27.73	±19.18	±15.94	±16.11	±14.83	±14.57
	σ gm.	228.91±19.61	179.84±13.56	139.80±11.27	139.29±11.39	160.10±10.49	133.12±10.30
	C. of V. per cent.	26.02± 2.38	13.61± 1.05	17.19± 1.43	10.90± .90	22.07± 1.52	12.69± 1.00
13	P. E. of mean gm.	±29.87	±16.85	±17.84	±17.52	±15.83	±17.16
	σ gm.	246.57±21.12	157.96±11.91	156.46±12.61	151.43±12.39	170.86±11.19	156.84±12.13
	C. of V. per cent.	23.42± 2.37	10.71± .82	15.98± 1.32	10.40± .86	20.23± 1.38	13.82± 1.09
14	P. E. of mean gm.	±32.30	±19.62	±18.55	±19.75	±16.19	±18.61
	σ gm.	286.58±22.83	183.96±13.87	162.69±13.12	170.73±13.96	174.71±11.45	170.12±13.16
	C. of V. per cent.	21.59± 1.93	11.30± .86	14.73± 1.21	10.62± .88	18.07± 1.22	13.48± 1.06
18	P. E. of mean gm.	±38.92	±23.66	±23.88	±27.36	±17.16	±24.38
	σ gm.	321.27±27.52	221.85±16.73	209.50±16.89	236.53±19.35	185.20±12.13	222.80±17.24
	C. of V. per cent.	18.21± 1.61	10.62± .81	12.49± 1.02	10.75± .89	13.46± .90	13.55± 1.07
22	P. E. of mean gm.	±37.70	±25.42	±30.16	±33.97	±18.46	±28.82
	σ gm.	311.14±26.65	238.30±17.97	264.50±21.32	293.63±24.02	199.24±13.05	263.40±20.38
	C. of V. per cent.	13.68± 1.19	9.64± .73	11.43± .93	11.31± .94	11.77± .78	13.72± 1.08
26	P. E. of mean gm.	±43.81	±27.20	±35.76	±39.77	±22.29	±32.35
	σ gm.	361.63±30.98	255.00±19.23	313.69±25.29	343.73±28.12	240.61±15.76	295.60±22.87
	C. of V. per cent.	13.36± 1.17	9.11± .69	11.79± .96	12.15± 1.01	11.97± .80	13.83± 1.09
30	P. E. of mean gm.	±48.49	±31.80	±41.44	±41.24		
	σ gm.	400.24±34.28	298.17±22.49	363.45±29.30	356.49±29.16		
	C. of V. per cent.	13.12± 1.14	9.68± .73	12.34± 1.01	11.43± .95		
34	P. E. of mean gm.	±46.53	±31.41	±48.88	±42.46		
	σ gm.	384.01±32.90	294.46±22.21	428.68±34.56	367.05±30.02		
	C. of V. per cent.	11.56± 1.00	8.57± .65	13.52± 1.11	11.00± .91		
38	P. E. of mean gm.	±	±	±55.53	±49.66		
	σ gm.			487.07±39.27	429.26±35.11		
	C. of V. per cent.			14.09± 1.16	12.07± 1.00		

After these data had been assembled it seemed desirable to compare the growth of the several lots of Rhode Island Red chicks with the growth of chicks of the same breed as published by other investigators. A search of the literature revealed the fact that there were but few instances in which complete, or even approximately complete, growth data had been obtained and published for this breed.

In 1918 Card and Kirkpatrick (*11*) [2] reported growth data for four lots of Rhode Island Red chicks from which the cockerels were removed on the twelfth, sixteenth, thirteenth, and twelfth weeks, respectively. Three years later Brody (*4*) used the average growth data of these four lots in a study of the rate of growth of the domestic fowl. Robertson (*28*) in turn, a few years later, cited Brody's analysis in support of his contention that the growth of animals may be

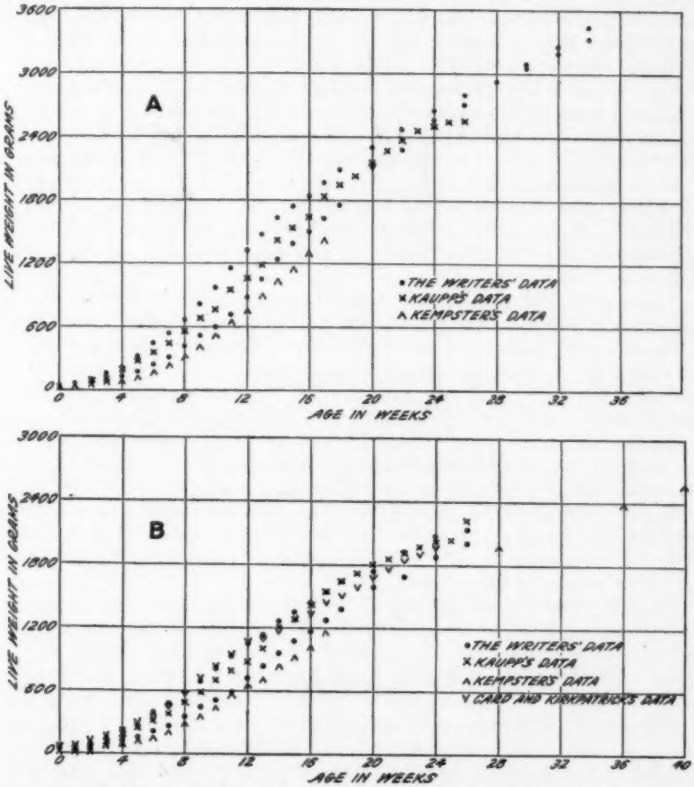

FIG. 1.—Comparison of the writers' growth data for Rhode Island Red chickens with the growth data obtained by other investigators for the same breed. A, cockerels; B, pullets. In each case the upper series of dots (.) represents the growth of the lot which received skim milk while the lower series of dots is for the lot which did not

represented by the formula for a monomolecular, autocatalytic, chemical reaction. In the same year (1923) in which Robertson's monograph appeared Kempster and Henderson (*16*) compared these data obtained by Card and Kirkpatrick with some published by Kaupp (*15*) in 1921.

Kaupp reported in 1921 the average growth of several lots of Rhode Island Red chicks by sexes. In obtaining his data he fed

[2] Reference is made by number (italic) to "Literature cited," p. 539.

similar rations to the several lots, the difference between the rations being that they contained different protein supplements, i. e., milk, meat scraps, and tankage.

In 1926 Kempster and Seaton (*17*) reported the growth, by sexes, of Rhode Island Red chicks up to the age of 17 weeks.

The writers plotted the several sets of data mentioned above with the data in Figure 1, A and B. In the case of the cockerels the average growth of Kaupp's several lots falls between the growth of the writers' lot receiving skim milk and the one not receiving it. The growth of Kempster's chicks is appreciably less than that of the writers' chicks which received no milk.

In the case of the pullets the average growth of Kaupp's chicks falls between that of the writers' two lots until the sixteenth week, after which it is similar to that of the writers' lot receiving milk. Again the growth of Kempster's chicks is less than that of those receiving no milk in the present investigation. For the first 12 weeks the growth of Card and Kirkpatrick's chicks (males and females) was very similar to the growth of the writers' pullets, but after that time the growth of their chicks (females) fell between that of the writers' two lots.

In general, the growth of cockerels and pullets in the studies here reported appears to be more nearly uniform than the growth of the cockerels and pullets of the other investigators; also, for the ages plotted, the writers' cockerels receiving skim milk were heavier than any of the others, and the pullets receiving skim milk were as heavy as the heaviest of either Kaupp's or Card and Kirkpatrick's pullets.

Inasmuch as a comparison of the writers' data with the data secured by other investigators indicates the great difficulty, if not impossibility, of setting up a "normal growth curve" for a given breed of chickens, the data given here are submitted, not as representing the "normal growth" of the Rhode Island Red breed but rather as representing the growth which was observed by the writers for their strain of that breed.

To show how the variability of the live weight of the chicks changed with increasing age, the coefficient of variation of the live weight of the chicks of each lot was plotted against age in Figure 2. (See Table 2.) In each case the resulting curve shows a characteristic hook shape (⌒——). For the three lots receiving skim milk (lot 2 cockerels, lot 2 capons, and lot 2 pullets) the period of high variability in live weight is confined to the first seven or eight weeks, whereas for the three lots not receiving skim milk (lot 1 cockerels, lot 1 capons, and lot 1 pullets) the period of high variability extends to at least the twelfth week.

Jull (*14*) has called attention to this increase in variability in live weight during the first four or five weeks in the case of Barred Plymouth Rock chicks. He found that after this period of high variability, which lasted until the tenth week for males and twelfth week for females, there was a general tendency toward decrease in variability in live weight for both sexes. In the case of each of the writers' lots of chicks the variability also showed a general tendency to decrease with age, and after the twenty-second week lay well between 7.5 and 15 per cent.

In regard to the relationship between sex and variability in live weight, the writers' data show that for the three lots (cockerels,

capons, and pullets) receiving skim milk the period of high variability, as well as the extent of the variation during this period, is very much the same; however, in the case of the three lots receiving no skim milk the period of high variability is about the same, but the extent of the variation during this period is much greater for the cockerels than the pullets and much greater for the pullets than the capons. No significance is attached to these differences in extent of variability because, until the age of 10 weeks, the chicks designated as capons were cockerels and it was before this age that the difference between lot 1 cockerels and lot 1 capons became most pronounced.

In many nutrition experiments with poultry growth is the main criterion for judging and analyzing the results. When the Rhode Island Red is used the writers' determinations of the probable error of the mean live weight for the different ages should give some indication of the probable error of the mean live weight to be expected

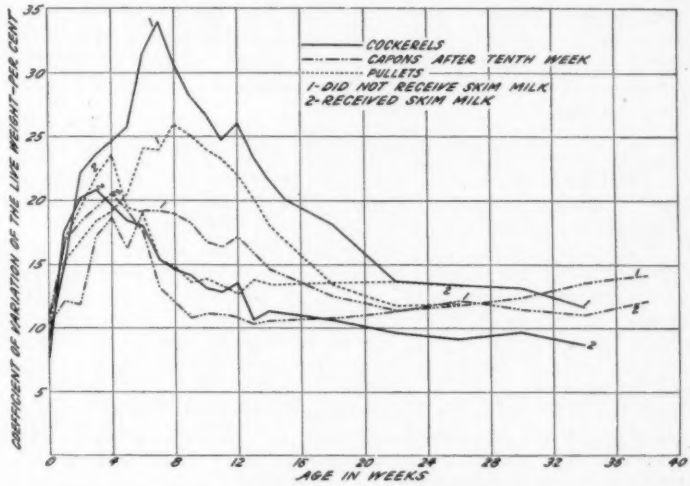

FIG. 2.—The variability of live weight with age

for normal lots of from 30 to 50 chicks. There is but one other breed, the Barred Plymouth Rock, known to the writers, for which the probable error of the mean live weight at different ages may be found in the literature; Jull (*14*) gives the probable error of the mean live weight of lots containing 38 of each sex of this breed for the first 22 weeks.

In a recent contribution to the study of growth Brody (*6*) calls attention to the sharp breaks frequently observed when time rates of growth are plotted and suggests that these are breaks between successive stages of constant growth rates, i. e., between successive cycles of growth. In the course of analyzing the present data the writers plotted the standard deviation of the weights of the chicks in each lot against time (age) and found analogous breaks. In many instances the plotted points fell approximately along a straight line for a period of several weeks and then there was a break and again

the plotted points fell approximately along a straight line; in the case of each lot of chicks there were four or five periods in which the plotted points fell approximately along straight lines. The standard deviation, when plotted against age, may give valuable clews as to the approximate time of the beginning and end of cycles of growth.

THE GROWTH CURVES

Many more or less successful attempts have been made to describe the growth of animals, as well as of plants, by means of mathematical formulas. Some investigators have been content to use wholly empirical formulas while others have attempted to derive rational ones. Robertson (*28*) has called attention to the similarity in shape of the curve of the monomolecular, autocatalyzed, chemical reaction and the curve of normal growth of animals and has attempted to describe the growth of animals (and plants) by means of the equation of this curve. Brody (*5*, *7*), observing that the successive periodic gains in the live weight of an animal seemed to bear a definite relationship to each other during the later stages of growth, employed the curve of diminishing increment in analyzing the growth of several species of animals.

It is not the present purpose to review the large amount of work which has been done in this field or to discuss the relative value of the several formulas which have been proposed by different investigators. Persons interested in this phase of the subject are referred to the numerous papers of Robertson and of Brody.

In the present study the writers selected the curves used by the above-mentioned investigators because those curves had been used in more instances than any others for describing the growth of chickens. The autocatalytic formula was selected for describing the average growth of the lots of chicks during the first 15 to 18 weeks and the formula of the curve of diminishing increment for the remainder of the growth.

In the course of the curve fitting the writers found that the autocatalytic curve did not describe the growth of the lots of chicks so well as had been hoped. So far as the writers' data are concerned, the autocatalytic formula is only an approximation for a part of the growth curve. This is in agreement with the contention of Van de Sande-Bakhuyzen (*29*, *30*) that the autocatalytic formula is only a first approximation for a part of the growth curve. It may also be remarked that Murray (*22*) has raised a number of objections to the autocatalytic concept of growth. However, despite the objections that have been raised against the autocatalytic formula for describing growth and the fact that the autocatalytic curve did not describe the average growth of the lots of chicks so well as had been hoped, the writers feel that the constants which were obtained in fitting this curve to the sets of data are of significance, at least empirically.

The curve of diminishing increment fully met the writers' expectations as to its suitability for describing the later stages of the growth of the six lots of chickens.

Before reporting the results of the curve fitting, a short description of the method used may be of interest. First, the constants for each curve, for each lot of chicks, were approximately determined by a combination of graphical and least square methods described by

Robertson (*28*) and by Brody (*5*). In the case of the autocatalytic curve $\left(\log \frac{w}{A-w} = K\,(t-t_1)\right.$,

in which

w = weight of the animal at any time t,
A = weight which the animal approaches asymptotically as the cycle of growth involved approaches completion.
K = the velocity constant of growth,
t = age, and
t_1 = age at which the cycle is half completed, i. e., when $w = \frac{A}{2}\Big)$,

the accuracy of the determination of the constants K and t_1 depends on the accuracy with which the constant A has been determined graphically; and in the case of the curve of diminishing increment [3]

$$\left(w = A - Be^{-k(t+3)},\right.$$

in which

w = weight of the animal at any time t,
A = mature weight of animal which is approached asymptotically as t approaches infinity,
B = a constant upon which the gain in weight of the animal is dependent and which in turn (as we believe) is dependent upon the feed which the animal receives as well as the kind of animal,
e = the base of the natural system of logarithms,
k = a constant indicating the rate of decline in growth of the animal,
t = age from time of hatching (birth),
3 = number of weeks required for hatching (period of gestation).$\Big)$

the accuracy of the determination of the constants B and k depends on the accuracy with which the constant A has been determined graphically.

Desiring extreme accuracy in the determination of the constants, the writers corrected them in the manner described by Robertson (*28*) in the appendix of his monograph on the chemical basis of growth and senescence. Finding that in most cases these corrections ranged from a fraction of a per cent to as much as 10 per cent, the writers continued to correct the "corrected" constants until negligible corrections were obtained.

Inasmuch as the writers' "correction" or adjustment equation was not the same as the one in Robertson's monograph, there is given here

[3] The curve of diminishing increment is sometimes written in the form, $w = A - BR^t$, or in the present case: $w = A - BR^{(t+3)}$, in which w, A, B, t, and 3 have the same significance as in the other form and $R = e^{-k}$. In this form of the equation the increase in weight during any period of time (week, if t is measured in weeks, etc.) is R (always a fraction) times that of the preceding period.

the adjustment equation used with each type of curve. For the autocatalytic equation the writers used:[4]

$$\frac{d\phi}{d\mathrm{A}}\alpha+\frac{d\phi}{d\mathrm{K}}\beta+\frac{d\phi}{d_1}\gamma=\theta$$

in which

$$\frac{d\rho}{d\mathrm{A}}=\frac{x}{\mathrm{A}},$$

$$\frac{d\phi}{d\mathrm{K}}-(t-t_1)\,\mathrm{A}V,$$

$$\frac{d\phi}{dt_1}=-\mathrm{K}\mathrm{A}V, \text{ and}$$

$$V=\frac{2.302585\cdot 10^{\mathrm{K}(t-t_1)}}{[1+10^{\mathrm{K}(t-t_1)2}]}.$$

In Robertson's adjustment equation[5] $\frac{d\phi}{d\mathrm{K}}=t\,\mathrm{A}V$, while in the writers' $\frac{d\rho}{d\mathrm{K}}=(t-t_1)\,\mathrm{A}V$.

For the curve of diminishing increment the writers used:[6]

$$\frac{d\phi}{d\mathrm{A}}\alpha+\frac{d\phi}{d\mathrm{B}}\beta+\frac{d\phi}{d\mathrm{k}}\gamma=\theta$$

in which:

$$\frac{d\phi}{d\mathrm{A}}=1,$$

$$\frac{d\phi}{d\mathrm{B}}=-\epsilon^{-\mathrm{k}(t+3)} \text{ or } -\frac{1}{\epsilon^{\mathrm{k}(t+3)}}, \text{ and}$$

$$\frac{d\phi}{d\mathrm{k}}=(t+3)\,\mathrm{B}\epsilon^{-\mathrm{k}(t+3)}$$

The constants for each curve, for each lot of chicks, as finally determined are given in Tables 5 and 6.

TABLE 3.—*Constants of the autocatalytic curve*[a] *fitted to the first 15 to 18 weeks of growth*

Constants	Cockerels		Capons		Pullets	
	Lot 1 (0–18 weeks)	Lot 2 (0–16 weeks)	Lot 1 (0–18 weeks)	Lot 2 (0–15 weeks)	Lot 1 (0–15 weeks)	Lot 2 (0–16 weeks)
A (grams)	2,234	2,133	2,250	2,090	1,747	1,599
K	0.1238	0.1414	0.1141	0.1412	0.1179	0.1390
t_1 (weeks)	13.40	10.46	14.06	10.43	13.27	9.87
S[b] (grams)	±16.66	±18.70	±13.25	±19.16	±8.79	±19.27

[a] $\log\frac{y}{\mathrm{A}-y}=\mathrm{K}(t-t_1)$

[b] S=root-mean-square deviation of the observed weights from the calculated weights.

[4] Employing Robertson's system of notation (*28*).
[5] Probably due to a typographical error.
[6] Employing Robertson's system of notation (*28*).

TABLE 4.—*Constants of the curve of diminishing increment* [a] *fitted to the later stages of growth*

Constants	Cockerels		Capons		Pullets	
	Lot 1 (16–34 weeks)	Lot 2 (14–32 weeks)	Lot 1 (14–38 weeks)	Lot 2 (13–38 weeks)	Lot 1 (12–26 weeks)	Lot 2 (11–26 weeks)
A (grams)	4,480	4,438	4,035	4,063	3,385	3,316
B (grams)	8,308	6,086	8,992	6,950	5,388	4,682
k	0.0538	0.0452	0.0650	0.0612	0.0471	0.0482
t^* [b] (weeks)	8.48	3.96	9.32	5.76	6.87	4.15
R [c]	0.9477	0.9551	0.9370	0.9406	0.9540	0.9529
S [d] (grams)	±29.74	±12.08	±42.78	±36.51	±14.13	±17.95

[a] $w = A - Bc^{-k(t+3)}$ or $w = A - BR^{(t+3)}$

[b] t^* = age at which $w = 0$. See Brody (5, 7).

[c] $R = e^{-k}$, i. e., R is the ratio of the gain in live weight during any week to that during the preceding week.

[d] S = root-mean-square-deviation of the observed weights from the calculated weights.

Having determined the constants of the two curves for each lot of chicks, the writers calculated the weights for each week and plotted the curves together with the observed weights. (Figs. 3, A; 4, A; 5, A; 6, A; 7, A; 8, A.) The calculated weights are given in Tables 5 and 6.

TABLE 5.—*Weights of the chicks for the first 15 to 18 weeks of growth, calculated by means of the formula for the autocatalytic curve*

Age (weeks)	Weight of—					
	Cockerels		Capons		Pullets	
	Lot 1	Lot 2	Lot 1	Lot 2	Lot 1	Lot 2
	Grams	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*
0	48	68	55	68	46	65
1	63	94	70	93	60	88
2	83	128	91	127	78	119
3	110	173	117	171	101	160
4	143	232	149	230	131	212
5	187	308	190	306	168	278
6	242	405	242	401	213	359
7	310	522	304	517	270	456
8	394	661	380	653	338	567
9	496	818	471	807	418	699
10	614	987	576	973	510	816
11	749	1,160	695	1,143	613	943
12	897	1,329	827	1,308	725	1,062
13	1,053	1,484	969	1,459	842	1,170
14	1,212	1,621	1,115	1,593	960	1,263
15	1,367	1,737	1,262	1,706	1,075	1,340
16	1,512	1,832	1,405	----	----	1,402
17	1,644	----	1,538	----	----	----
18	1,759	----	1,660	----	----	----

In order to visualize better the average growth of the six lots of chicks, the writers plotted the observed average weekly gains against age, as well as the smoothed [7] average weekly gains and the calculated average weekly gains.[8] (Figs. 3, B; 4, B; 5, B; 6, B; 7, B; and 8, B.) In this way some interesting points were brought out which will be discussed later.

[7] Method of moving averages.

[8] Not the instantaneous rates of gain.

TABLE 6.—*Weights of the chicks for the later stages of growth, calculated by means of the formula for the curve of diminishing increment* [a]

Age (weeks)	Weight of—					
	Cockerels		Capons		Pullets	
	Lot 1	Lot 2	Lot 1	Lot 2	Lot 1	Lot 2
	Grams	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*
11					(597)	933
12					725	1,045
13			(858)	1,454	847	1,152
14	(1,149)	1,618	1,058	1,609	964	1,254
15	(1,323)	1,743	1,246	1,755	1,075	1,351
16	1,489	1,862	1,421	1,892	1,182	1,444
17	1,645	1,976	1,586	2,021	1,283	1,532
18	1,793	2,085	1,740	2,143	1,379	1,616
19	1,934	2,189	1,884	2,257	1,472	1,696
20	2,067	2,288	2,020	2,364	1,560	1,772
21	2,194	2,383	2,147	2,465	1,644	1,845
22	2,313	2,474	2,266	2,560	1,724	1,914
23	2,427	2,561	2,377	2,649	1,800	1,980
24	2,534	2,644	2,481	2,733	1,873	2,043
25	2,636	2,723	2,579	2,812	1,942	2,103
26	2,733	2,799	2,671	2,886	2,009	2,160
27	2,824	2,872	2,757	2,956	(2,072)	(2,214)
28	2,911	2,941	2,837	3,022	(2,132)	(2,266)
29	2,993	3,007	2,913	3,084	(2,190)	(2,316)
30	3,071	3,071	2,983	3,142	(2,245)	(2,363)
31	3,144	3,131	3,049	3,197	(2,297)	(2,408)
32	3,214	3,189	3,112	3,248	(2,347)	(2,451)
33	3,281	(3,244)	3,170	3,297	(2,395)	(2,491)
34	3,343	(3,297)	3,224	3,342	(2,440)	(2,530)
35	(3,403)	(3,347)	3,275	3,385		
36	(3,459)	(3,396)	3,323	3,425	(2,525)	(2,602)
37	(3,513)	(3,442)	3,368	3,463		
38	(3,563)	(3,486)	3,410	3,499	(2,603)	(2,668)
39	(3,611)	(3,528)	(3,449)	(3,532)		
40	(3,657)	(3,568)	(3,486)	(3,564)	(2,673)	(2,728)

[a] The weights in parentheses are extrapolated values; i. e., they lie outside the age intervals to which the curves were fitted.

A casual inspection of Figures 3 to 8, inclusive, leaves an impression that, in general, the curves selected to describe the growth of the chicks fulfilled this purpose rather well. As a comparison of the observed with the calculated weights (Tables 1 and 6) shows, this is true in the case of the curve of diminishing increment; however, in the case of the autocatalytic curve the agreement between the observed and calculated weights is not so good for the first several weeks, except, possibly, in the case of lot 1 cockerels and lot 1 pullets. (Compare Tables 1 and 5.)

This is contrary to the results obtained by Brody (*4*) in fitting this curve to Card and Kirkpatrick's (*11*) growth data for the Rhode Island Red chicken, for in this instance the agreement between the observed and calculated weights was unusually close. Since environmental, as well as nutritional, conditions are such potent factors in the early growth of chickens, it seems almost too much to expect that any one type of curve would be able to describe their early growth unless these conditions were rigidly controlled. It is therefore to be anticipated that there will be cases in which a given type of curve will accurately describe the first several weeks of the growth of chickens as well as cases in which it will fail to do so.

A study of Tables 3 and 4 shows that in each case the constants of the growth curves were markedly affected by allowing the birds

free access to sour skim milk in addition to feeding them the basal ration. In the case of the constants of the autocatalytic curve, the constant A[9] was depressed 4.5 per cent (cockerels) to 8.5 per cent

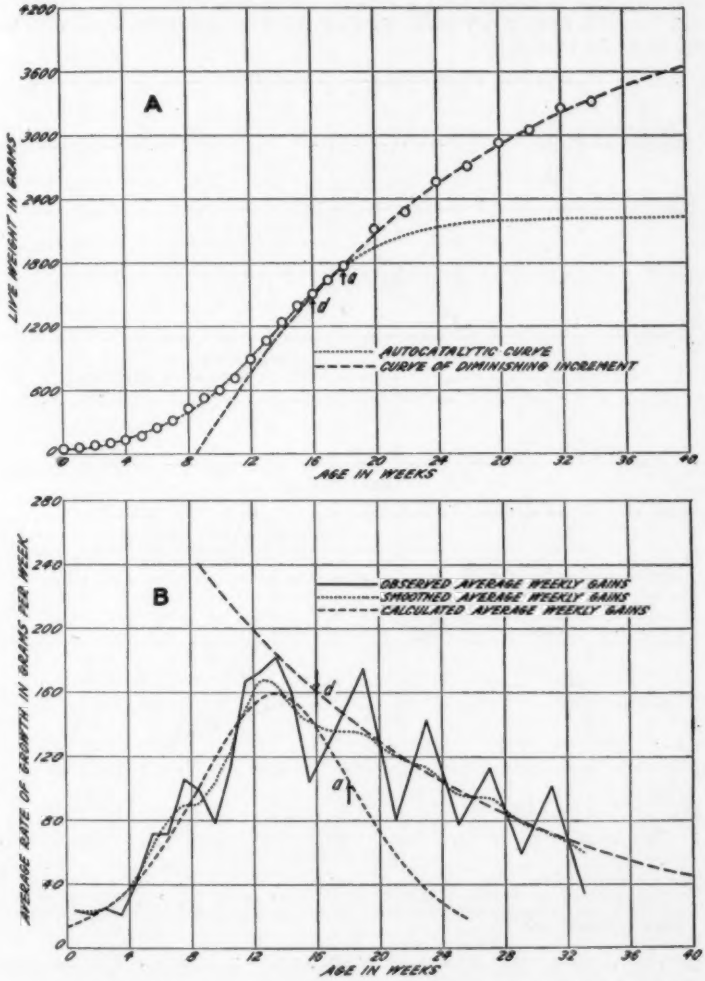

FIG. 3.—Lot 1 cockerels. A, growth curves fitted to the observed average weights; B, average-rate-of-growth curves. In each case *a* marks the end of the age interval to which the autocatalytic curve was fitted and *d* marks the beginning of the age interval to which the curve of diminishing increment was fitted

(pullets), but the velocity constant of growth K was increased 13.4 per cent (cockerels) to 23.76 per cent (capons), and the time t_1 required to make one-half of the growth indicated by constant A

[9] For the explanation of the constants discussed, see p. 523.

was decreased 21.9 per cent (cockerels) to 25.8 per cent (capons). That is, the feeding of sour skim milk slightly decreased the amount of growth indicated by the constant A but very greatly decreased the time required to make that growth. In brief, then, it may be stated that feeding sour skim milk greatly accelerated growth during the first 15 to 18 weeks.

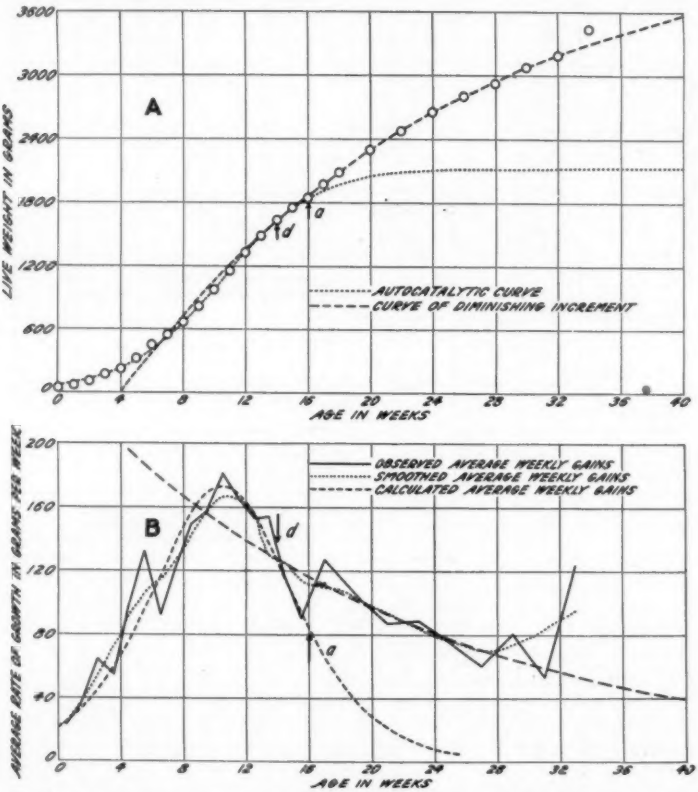

FIG. 4.—Lot 2 cockerels. A, growth curves fitted to the observed average weights; B, average-rate-of-growth curves. In each case *a* marks the end of the age interval to which the autocatalytic curve was fitted and *d* marks the beginning of the interval to which the curve of diminishing increment was fitted

In the case of the curve of diminishing increment the average mature weight of the chickens, as represented by the constant A, was not greatly affected, but the constant B, upon which the magnitude of the absolute weekly gains is dependent, was very greatly decreased. As for k, considered by itself, it seems that the feeding of sour skim milk had no consistent effect on it. In each case t^* was greatly decreased in value. R appears to have remained rather constant not only for the different nutritional conditions but also for the different sexes; this is to be expected from the relationship between

R and k, rather large changes in k corresponding to rather small changes (percentage basis) in *R*.

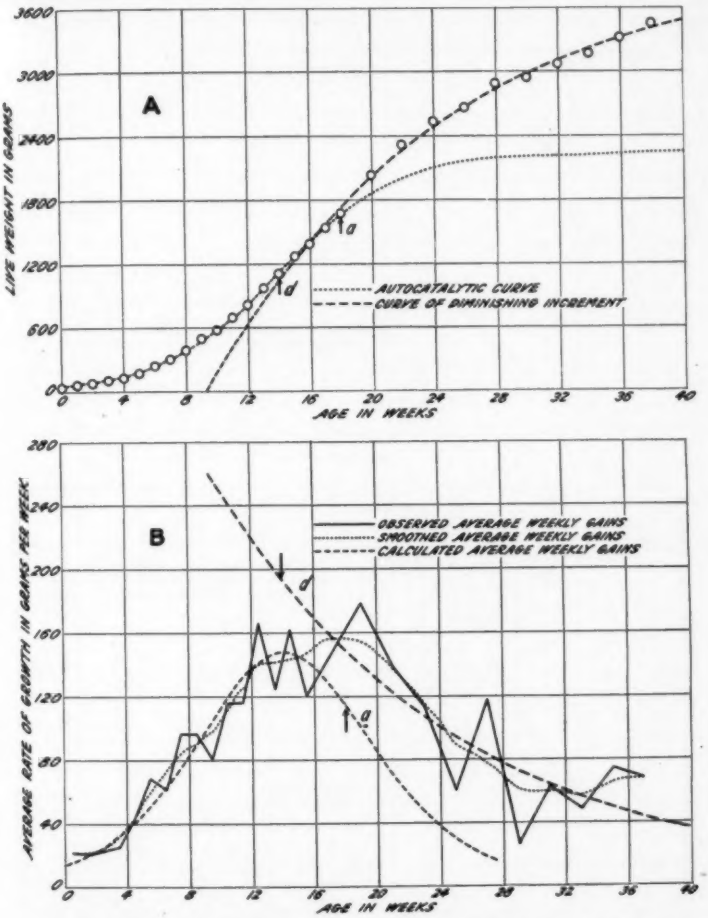

FIG. 5.—Lot 1 capons. A, growth curves fitted to the observed average weights; B, average-rate-of-growth curves. In each case *a* marks the end of the age interval to which the autocatalytic curve was fitted and *d* marks the beginning of the interval to which the curve of diminishing increment was fitted

GENERAL DISCUSSION

THE EFFECT OF MILK ON GROWTH OF CHICKENS

Investigators for at least the last 35 years have loudly praised the value of milk (usually skim milk [10]) for feeding poultry. In 1898

[10] According to Card (*10*), buttermilk and sour skim milk have approximately the same feeding value. Except in extreme cases, it is probable that all the forms of milk usually fed to poultry have approximately the same feeding value.

Anderson (*1*) concluded that skim milk was especially valuable as a food for young chickens, but became of less importance as the chickens grew older. After repeating his experiments (*2*) he concluded that skim milk did not decrease in value as the chickens became older.

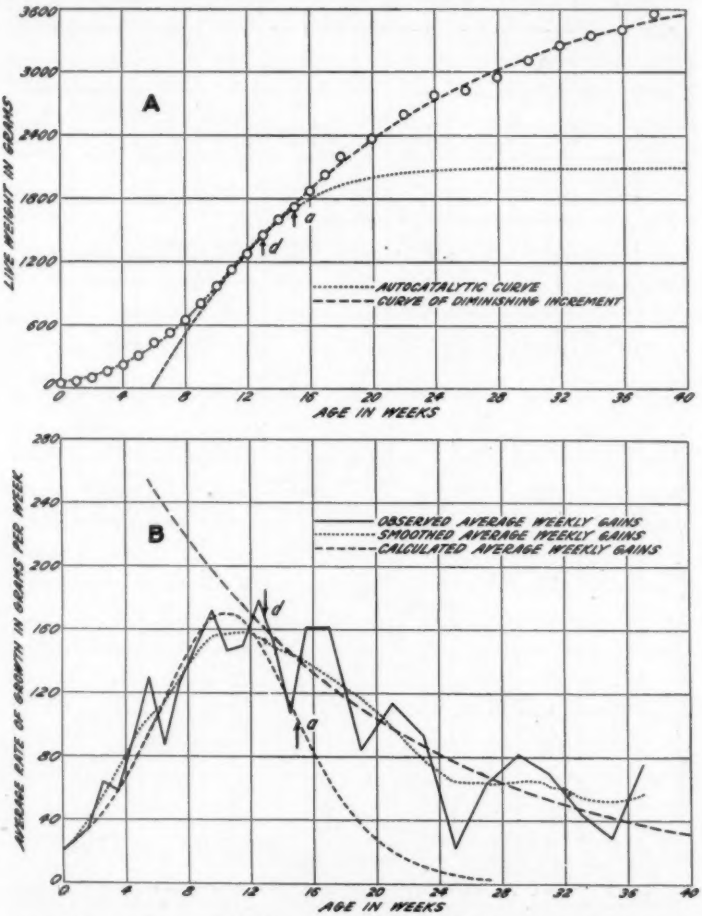

FIG. 6.—Lot 2 capons. A, growth curves fitted to the observed average weights; B, average-rate-of-growth curves. In each case *a* marks the end of the age interval to which the autocatalytic curve was fitted and *d* marks the beginning of the interval to which the curve of diminishing increment was fitted

It seems to the writers that the value of skim milk, at any time, would depend to a great extent on the feed fed with it. The writers' results fit his first conclusion regarding age rather than his second. The basal ration which he fed the second year was not quite the same as that fed the first year and probably not so adequate for growth.

Wheeler (*32*) made a comparison of rations wholly of vegetable origin and others containing feed of animal origin, and from his results concluded that after the period of most active growth had passed and the young chickens were approaching maturity the difference in efficiency between such rations rapidly disappeared. His results parallel those of the writers.

Previous to Anderson's experiments, work done at the New York station (*23*) indicated the generally beneficial effect of skim milk on health and mortality. Rettger, Kirkpatrick, and Card (*27*) in 1915

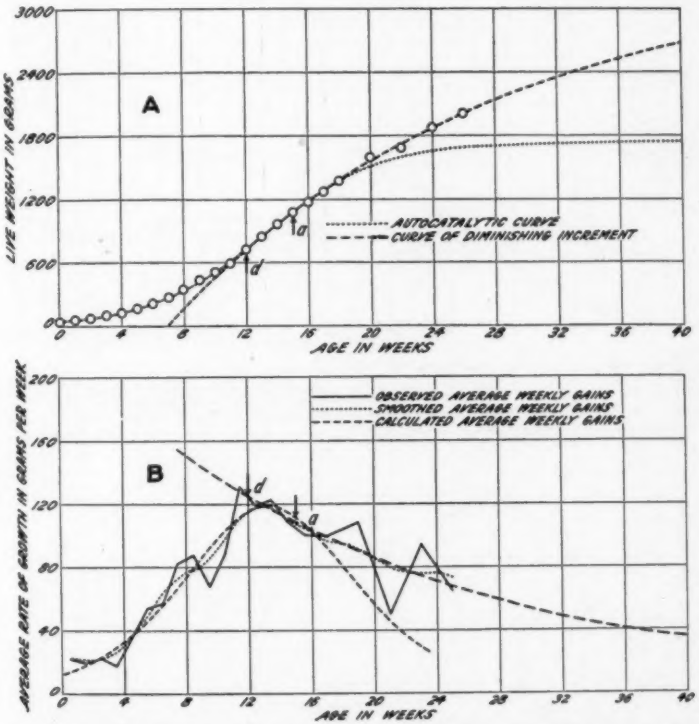

FIG. 7.—Lot 1 pullets. A, growth curves fitted to the observed average weights; B, average-rate-of-growth curves. In each case *a* marks the end of the age interval to which the autocatalytic curve was fitted and *d* marks the beginning of the interval to which the curve of diminishing increment was fitted

stated that it was most conclusively shown in all their experiments that milk feeding stimulated growth and caused a great reduction in deaths from general causes. Since then a number of investigators, Goodale (*13*), Lewis (*19*), Orr (*24*), and others, have testified to the beneficial effect of milk on growth.

Considering it as an established fact that milk does have a marked effect on the growth of chickens, it is desirable to know how, when, and why this effect is manifested. In Figures 9, A and B, 10, A and B, and 11, A and B, the writers have plotted the growth curves and

the smoothed average-rate-of-growth curves of the cockerels, capons, and pullets to show the effect of feeding milk. In each of the three cases the cumulative growth curves diverge from each other after the first week, but after the twenty-eighth week they either come together again or else show a marked tendency to do so. Likewise, in each of the three cases, the smoothed average-rate-of-growth curves separate at the start but in this instance they come together again and *cross* at about 12 weeks for the cockerels, 14.5 weeks for capons, and 11.5 weeks for pullets.

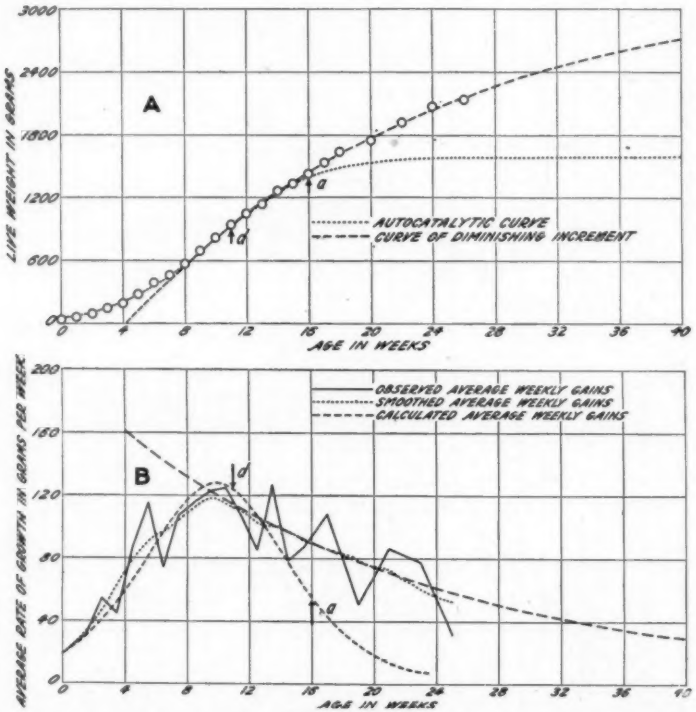

FIG. 8.—Lot 2 pullets. A, growth curves fitted to the observed average weights; B, average-rate-of-growth curves. In each case *a* marks the end of the age interval to which the autocatalytic curve was fitted and *d* marks the beginning of the interval to which the curve of diminishing increment was fitted

On examining the calculated average-rate-of-growth curves for the first 15 to 18 weeks (figs. 3 to 8, inclusive) one finds that in each case the curve for the lots receiving skim milk is higher and narrower than for the lots not receiving it. It is clear, then, according to the data, that the effect of skim milk on growth is shown by an increased rate of growth during the first 12 to 15 weeks.

Regarding the cause of this effect there is much speculation and uncertainty. In general, the cause is ascribed to the mineral and protein content of the milk, particularly the latter. Buckner,

Nollau, Wilkins, and Kastle (*8*) believed that the deficiency of certain grain rations for growth was due to the quality of the protein. In an earlier contribution of Buckner, Wilkins, and Kastle (*9*) some experiments were reported which indicated that the lysine content of certain grain mixtures was the limiting factor for growth. Although these experiments were open to criticism, Osborne and Mendel (*25*) later in part corroborated their findings. It may be that milk is

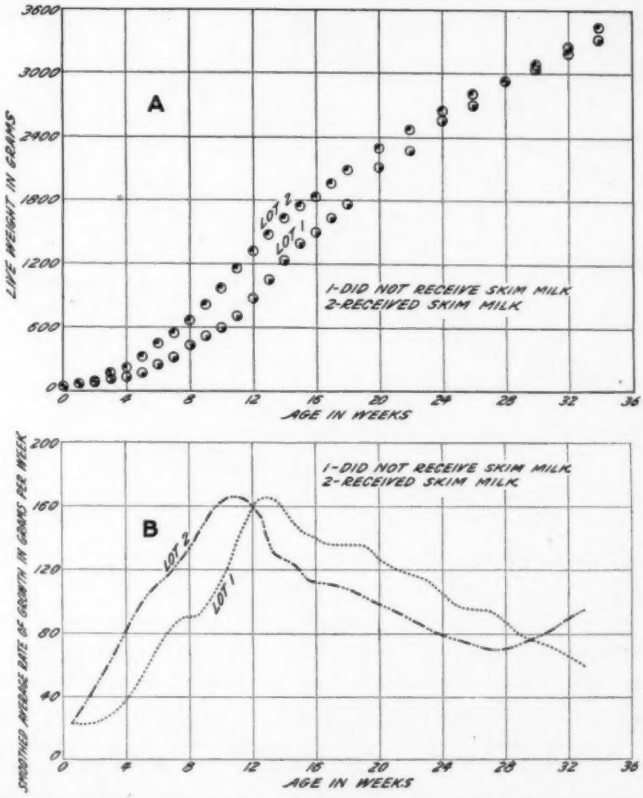

FIG. 9.—Comparison of the growth of the cockerels receiving skim milk with those not receiving it. A, cumulative growth curves; B, smoothed average-rate-of-growth curves

particularly valuable because of its proteins, and it is also possible that its value is due in part to its mineral content. One very striking thing is that feeding milk to chickens nearly always, if not always, increases the total feed consumption, and this in itself would tend to cause more rapid growth. However, it is easy to fall into the error of mistaking cause for effect, and if one assumes that milk is valuable because it increases feed consumption he is immediately called upon to explain why it does this.

Brody (7) has used the values of t^* (Table 4) for different animals (mammals) in comparing their equivalence of age. As the writers take it, he has assumed that t^* has a more or less fixed value for each kind of animal. According to the writers' values of t^*, this assumption as to constancy does not hold for Rhode Island Red chickens. However, if the hypothesis is applied to nutritionally different lots of chicks of the same breed, the writers' values of t^*[11] indicate that

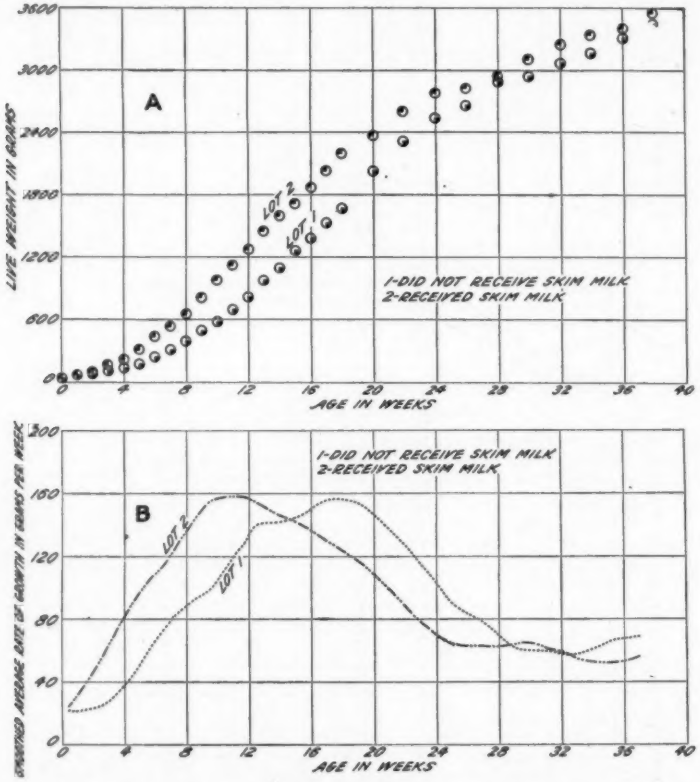

FIG. 10.—Comparison of the growth of the capons receiving skim milk with those not receiving it. A, cumulative growth curves; B, smoothed average-rate-of-growth curves

4 weeks of age in the cockerels receiving skim milk corresponded to 8.5 weeks of age in those not receiving it; that 5.8 weeks of age in the capons receiving skim milk corresponded to 9.3 weeks of age in those not receiving it, and that 4.2 weeks of age in the pullets receiving skim milk corresponded to 6.9 weeks of age in those not receiving it. It is to be noted that t^* for the capons was computed from data obtained after caponization and that the apparent discrepancy

[1] It should be noted that Brody's t^* is age from conception while the writers' is from birth (hatching).

between the differences in the two values for capons and for cockerels is probably due to this fact.

It is interesting to note that in the case of the lots of chickens receiving skim milk the point of inflection of the growth curve occurs earlier than in the case of those lots not receiving it; or, what appears to be the same thing, the whole of the growth curve of the former lots more nearly approaches the curve of diminishing increment than it does for the latter lots. Does this mean, then, that the more nearly a "perfect" condition of nutrition is reached the more nearly will the whole of the growth curve approach the curve of diminishing increment? This speculation is worthy of further consideration.

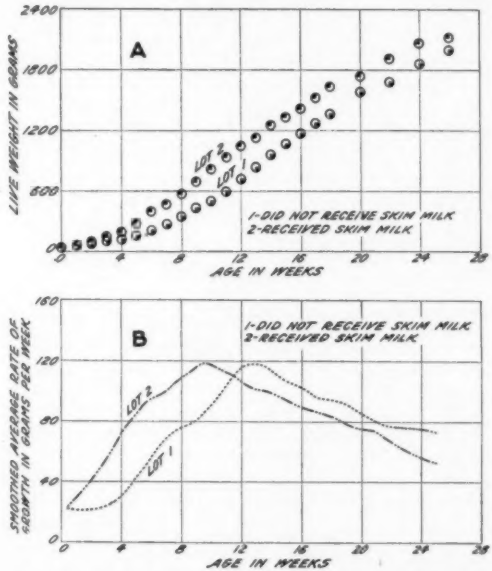

FIG. 11.—Comparison of the growth of the pullets receiving skim milk with those not receiving it. A, cumulative growth curves; B, smoothed average-rate-of-growth curves

THE INFLUENCE OF SEX ON THE GROWTH OF CHICKENS

It is a matter of common knowledge that the absolute rate of growth of the female chicken is less than that of the male and that its mature weight is less than that of the male. In the many records consulted the writers have not found a single exception to the above statement when the nutritional conditions were the same for both sexes. On this point the writers examined the growth data published by a number of investigators, including Bethke and Kennard (*3*), Buckner and his associates (*8*, *9*), Card and Kirkpatrick (*11*), Jull (*14*), Kaupp (*15*), Kempster (*16*, *17*), Latimer (*18*), Mitchell, Card, and Hamilton (*20*), and Philips (*26*).

In order to compare the rates of growth of the sexes, the writers have plotted together the smoothed average rates of growth of the

cockerels, capons, and pullets. (Fig. 12.) In the case of the lots receiving skim milk as well as in the case of the lots not receiving it, the rate of growth of the female is distinctly less than that of the male throughout the interval for which the writers obtained data on both sexes. According to the values of the constant A (Table 4) for the curve of diminishing increment, the mature female Rhode Island Red is on an average 1,108 gm. lighter than the male, or on a percentage basis, nearly 25 per cent lighter.

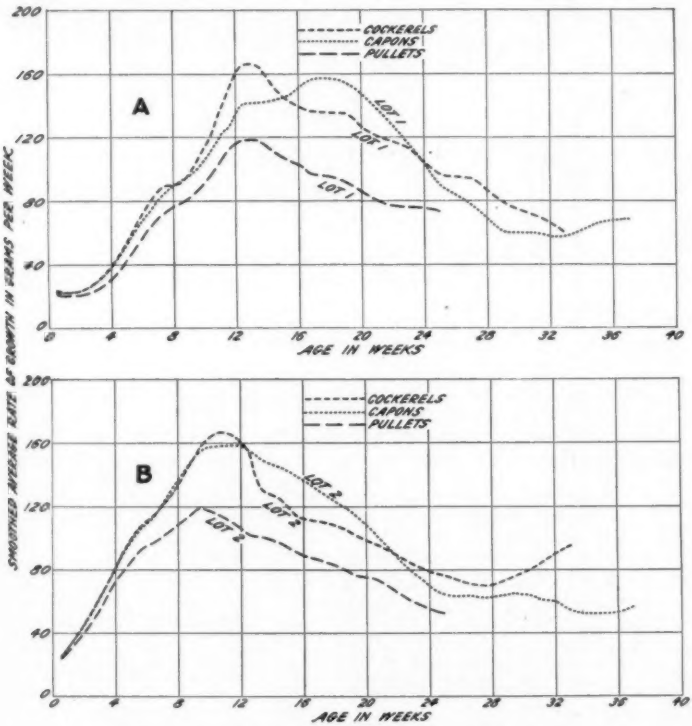

FIG. 12.—Comparison of the average-rate-of-growth curves for the different sexes. A, chickens not receiving skim milk; B, chickens receiving it

By means of the formula for the curve of diminishing increment one may calculate the time required for the two sexes to reach maturity (i. e., 98 per cent of the constant A, for, theoretically, A is never reached). Making the necessary calculations, the writers found that both the male and the female reach maturity (as defined above) at practically the same age, i. e., from 21 to 23 months. This is in fair agreement with the value obtained by Brody (*5*) for females of this breed, i. e., 25.2 months, or 25.9 months from the time the eggs were set.

COCKERELS VERSUS CAPONS

Waite (*31*) has called attention to the fact "that the opinion is quite general among those interested in poultry, that capons make a much more rapid growth and attain a size nearly twice that of cockerels of the same age and breed." In order to obtain some definite data on the question of the relative growth of capons and cockerels, he conducted a rather carefully controlled experiment. After analyzing his results he found that there was practically no difference until the cockerels began to reach maturity, at which time the capons made slightly better gains. The breed used was the White Plymouth Rock.

In 1918 Philips (*26*) reported the results of a study on the cost of raising White Plymouth Rocks. He stated that capons and cockerels grew with similar rapidity and retained similar weights until they reached 6½ pounds and that cockerels made gains at less cost per pound for feed than pullets or capons.

Elford (*12*) compared rates of fattening and economy of gains of cockerels and capons, using 22 Barred Rock cockerels and 22 Barred Rock capons. His conclusion was that the value of caponizing lies rather in the production of meat of superior quality than in any greater efficiency of growth or fattening.

Mitchell, Card, and Hamilton's (*20*) rather comprehensive study of the growth of White Plymouth Rock chickens furnishes some comparative data on the growth of cockerels and capons. They found that the growth of the capon was not distinctly different from that of the cockerel. In a popular report of their work (*21*) they state that "cockerels and capons grew at nearly the same rate up to an average weight of 6 pounds, when the growth records were discontinued."

A confusion of the terms "growth" and "fattening" seems to be responsible for the different opinions held on this question. Under the usual conditions of rearing cockerels and capons, the capon seems to have a much greater tendency to fatten than the cockerel. This means that eventually the capon may become heavier than the cockerel but, as intimated, this is due to its greater tendency to fatten and not to any greater ability to grow. In fact, the writers' data seem to indicate that the capon's ability to grow, i. e., in the strict sense of the word, is somewhat lessened by the operation of caponizing.

The writers' data (Table 1) show that, in the case of Rhode Island Reds, the average cumulative growth of the two lots of cockerels was very similar to that of the two corresponding lots of capons; if anything, the cumulative growth of the latter, from the time of caponizing up to 34 weeks of age, was a little less than that of the former. In Figure 12 the smoothed average rate of growth of the cockerels is compared with that of the capons. Up to the time of caponizing (10 weeks) the average rate of growth of the lots designated as lot 1 cockerels, and lot 1 capons was similar as was also that of the lots designated as lot 2 cockerels and lot 2 capons. Comparing the average rate of growth of these lots after the caponizing, one finds that in the case of the birds not receiving skim milk the average rate of growth of the cockerels was greater than that of the capons up to the age of 15 weeks, less between the fifteenth and twenty-

fourth weeks, and greater for at least the next 9 weeks. In the case of the birds receiving skim milk the average rate of growth between the thirteenth and the twenty-second week was greater for the capons than the cockerels, but less from the twenty-second to at least the thirty-third week.

Comparing the values of A (Table 4) for the curve of diminishing increment for each lot of cockerels and capons, one finds that they are appreciably greater for the former than for the latter. These values of A indicate that the weight of the Rhode Island Red cockerel, when it matures, is approximately 4,459 gm., while that of the Rhode Island Red capon is only about 4,049 gm. This does not mean that the capon may not become heavier than the cockerel, for it may easily do so, owing to its greater tendency to fatten.

As already stated, the time required for cockerels and pullets to reach maturity was found to be from about 21 to 23 months. Similar calculations for the two lots of capons show that the time required is approximately the same for both lots and is 17.4 months. This, then, is one point in favor of the capon, i. e., that it matures nearly 5 months earlier than the cockerel.

Mitchell, Card, and Hamilton (*21*) found that in the protein content of gains put on, the cockerels ranked above the capons. This is in agreement with the writers' conclusion that so far as growth in its stricter sense is concerned, the cockerel is, if anything, superior to the capon.

SUMMARY

Growth data were obtained on two groups of Rhode Island Red chicks which received the same treatment and the same feed, with the one exception that one group had access to both sour skim milk and water while the other group received only water. Each group was composed of one lot each of cockerels, capons, and pullets.

A study of the variability of the growth data suggested to the writers that the variability of live weight, as indicated by the standard deviation, may be of some value in determining the approximate time of the beginning and end of growth cycles.

A period of high variability in live weight was observed in the case of each of the six lots. For the lots receiving skim milk this period was between 0 and 7 or 8 weeks of age and for the lots not receiving skim milk it was between 0 and at least 12 weeks of age.

The curve of a monomolecular, autocatalyzed, chemical reaction was fitted to the growth made by each lot during the first 15 to 18 weeks and the curve of diminishing increment was fitted to the later stages of the growth of each lot.

Most of the constants of each curve were markedly affected by the feeding of sour skim milk.

Feeding sour skim milk had no marked effect on the calculated mature weights.

The data indicate that milk feeding accelerated growth during the first 15 to 18 weeks; according to the smoothed average-rate-of-growth curves, this acceleration took place between the time of the first feeding and 11.5 to 14.5 weeks of age.

The data further show that the rate of growth of the chicks receiving no skim milk was somewhat greater during the later stages of growth than that of the chicks receiving it.

The constant R (or $\epsilon - k$) for the curve of diminishing increment was not affected by feeding milk; its value was rather constant for each of the six lots.

The calculated average-rate-of-growth curves for the first 15 to 18 weeks were found to be narrower and higher for the lots receiving skim milk than for the lots not receiving it.

In the case of the lots of chicks receiving skim milk the point of inflection of the growth curve occurred earlier than in the case of the lots not receiving it. This suggested to the writers that the more nearly a "perfect" condition of nutrition is approached the more nearly will the whole of the growth curve approach the curve of diminishing increment.

The mature weights of Rhode Island Red cockerels, capons, and pullets were found to be, according to the values of the constant A for the curve of diminishing increment, 4,459, 4,049, and 3,351 gm., respectively.

By means of the equation of the curve of diminishing increment it was estimated that the ages at which maturity is reached are 21 to 23 months for cockerels and pullets and 17.4 months for capons.

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GROWTH OF CHICKENS IN RELATION TO FEED CONSUMPTION[1]

By MORLEY A. JULL, *Poultry Husbandman*, and HARRY W. TITUS, *Associate Biological Chemist, Bureau of Animal Industry, United States Department of Agriculture*

RELATION BETWEEN GROWTH AND FEED CONSUMPTION

The rate at which an animal producing flesh for human consumption gains in weight, as expressed by the number of units of feed required for the successive unit increases in live weight, is a problem of considerable practical importance. In the raising of poultry for market it is well recognized that beyond a certain stage in growth the value of the feed required to produce 1 pound gain in weight of live animal is out of all proportion to the value of the gain. Hence there has developed the common practice of marketing growing poultry at from 8 weeks to about 8 months of age. So far as the authors are aware, however, no experimental work has ever been undertaken to determine precisely the relationship between growth and feed consumption in the domestic fowl.

In poultry husbandry there are several types of phenomena which involve a gradual decrease in the per unit effect of some causal factor as the quantity of the factor acting increases. One phenomenon illustrating this point is the annual decrease in the egg production of hens, which after three or four years is so great that few hens produce eggs enough to justify the feed consumed. At the United States Animal Husbandry Experiment Farm, Beltsville, Md., it was found that for a period of three years, with a small group of birds, the annual average egg production per bird was 236, 188, and 167 for the first, second, and third years, respectively. Egg production decreased each year, and assuming that laying hens consume approximately the same amounts of feed each succeeding year, it is obvious that egg production would soon become unprofitable. Brody, Henderson, and Kempster,[2] in a rather large group of birds of two breeds, observed that each year's production was 88 per cent of that of the preceding year.

Another illustration of gradual decrease in the per unit effect of some causal factor as the quantity of the factor acting increases, may be cited in the results obtained at the United States Animal Husbandry Experiment Farm in feeding laying hens different quantities of protein. In rations in which the protein, except that contained in the grains, was provided exclusively from meat scraps, egg production increased as the protein level was raised, but above a protein level of 20 per cent egg production did not increase at the same rate as the protein level was increased. In other words, as the protein level was increased above the 20 per cent level there was relatively a decreasing rate of egg production.

[1] Received for publication Jan. 27, 1928; issued May 1, 1928.

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In the growing of chickens the same principle applies; there comes a time when the rate of growth decreases although feed consumption may be on the increase. Available data indicate that the relationship between increase in live weight in growing chickens and increase of feed consumed is expressed by the law of diminishing increment. Though comparatively few data are available to test the application of the law in the case of growing chickens, a rather complete discussion of it in its application to cattle, hogs, and chickens is given by Spillman.[3] A very interesting feature of the law is that it provides for the calculation of the rate of gain per feed unit, and the number of feed units per pound of gain, in growing chickens and in chickens that are being fattened for market. The purpose of the present experiment was to provide data, obtained under controlled conditions, to test the application of the law of diminishing increment as applied to growing chickens.

EXPERIMENTAL PROCEDURE

On April 24, 1925, there were hatched 170 chicks from a pen of Barred Plymouth Rock females mated to Rhode Island Red males. This mating was used in order to make possible the separation of the sexes at hatching time, there being involved the sex-linked barring factor of the females which was transmitted to the male chicks only. The male chicks had a white patch on the back of the head and also had yellow shanks, whereas the female chicks were without the white spot and had black shanks.

Of the 170 chicks, 84 were females and 86 were males. The females were divided into lot 1 with 40 chicks and lot 2 with 44 chicks. The males were divided into lot 3 with 43 chicks and lot 4 with 43 chicks. During the first three days there were 7 deaths in lot 1, 3 in lot 2, 0 in lot 3, and 3 in lot 4. Therefore, the number of chicks in each lot on which data were obtained was: Lot 1, 33 females; lot 2, 41 females; lot 3, 43 males; and lot 4, 40 males.

All lots were treated as uniformly as possible in respect to brooding and management. Each lot was brooded under an electric brooder in a pen 4 feet by 15 feet which adjoined a concrete yard 4 feet by 8 feet. At 8 weeks of age each lot was moved to a colony house 5 feet by 6 feet in a yard 100 feet by 200 feet and kept there until the conclusion of the experiment.

The four lots received the same ration and were fed in the same manner. During the first four weeks commercial chick feed was fed once daily and dry mash was fed twice daily, the mash being left before the chicks in hoppers for one hour each feeding time. After the first four weeks a scratch ration of 2 parts, by weight, cracked corn and 1 part wheat was fed twice daily and dry mash was kept in self-feeding hoppers before the birds at all times. The amount of scratch feed given daily was determined by the appetites of the birds, only such quantities being fed each day as would be eaten within a few minutes after being fed. The dry mash from

[3] SPILLMAN, W. J., and LANG, E. THE LAW OF DIMINISHING RETURNS. 178 p., illus. Chicago, World Book Co. 1924.

hatching time to the conclusion of the experiment was composed of the following parts by weight: Corn meal 40, sifted crushed oats 40, bran 20, middlings 20, beef scraps 10, dried buttermilk 10, bone meal 5, chick oyster shell 4, and cod-liver oil 3. The birds had free access to grit and water at all times.

Each chick was weighed separately at hatching time and every two weeks thereafter, the weights being recorded in grams to the second decimal place. The total amount of feed consumed by each lot was determined at the end of every two-week period.

The mortality throughout the duration of the experiment was comparatively low, one bird in each lot dying on the dates given: Lot 1, May 18 and July 17; lot 2, no deaths; lot 3, June 8 and 11, July 17 and 31; lot 4, May 20, July 16, and September 23.

AVERAGE WEIGHTS AND FEED CONSUMPTION

Table 1 gives the mean or average weight per chick in each of the four lots at hatching time and at the end of each two-week period thereafter. It will be observed that for the first 6 or 8 weeks the females weighed practically the same as the males, but that after 10 weeks the males increased in weight more rapidly than the females. A similar phenomenon was observed by Jull [4] in the case of Barred Plymouth Rock male and female chicks and by Titus and Jull [5] in the case of Rhode Island Red male and female chicks.

TABLE 1.—*The mean (average) weight per chick in each of the four lots at hatching time and each two-week period thereafter*

Age (in weeks)	Mean weight per chick in—				Age (in weeks)	Mean weight per chick in—			
	Lot 1 (females)	Lot 2 (females)	Lot 3 (males)	Lot 4 (males)		Lot 1 (females)	Lot 2 (females)	Lot 3 (males)	Lot 4 (males)
	Grams	*Grams*	*Grams*	*Grams*		*Grams*	*Grams*	*Grams*	*Grams*
0	34.54	34.19	34.97	34.77	14	1,334.55	1,236.59	1,505.87	1,629.74
2	90.82	79.76	77.12	82.27	16	1,541.29	1,480.37	1,772.95	1,933.03
4	215.31	198.10	185.44	225.59	18	1,651.77	1,574.15	2,046.03	2,157.37
6	387.25	355.76	366.77	399.43	20	1,860.16	1,785.43	2,386.79	2,435.53
8	618.13	582.71	608.10	656.59	22	2,026.93	2,012.07	2,736.41	2,744.86
10	835.94	770.85	773.17	924.23	24	2,211.45	2,170.97	2,906.28	2,926.89
12	985.45	936.95	1,091.10	1,225.66					

Table 2 gives the various statistical constants for the mean weights shown in Table 1. The values for the coefficients of variation show that the live weights of the birds in lot 1 (females) were most variable during the first two weeks after which they steadily became more uniform, but that in lots 2 (females), 3 (males), and 4 (males) the variability in live weight increased up to six weeks, decreasing almost steadily thereafter.

[4] JULL, M. A. DIFFERENTIAL SEX GROWTH CURVES IN BARRED PLYMOUTH ROCK CHICKS. Sci. Agr. 4: 58–65, illus. 1923.

[5] TITUS, H. W., and JULL, M. A. THE GROWTH OF RHODE ISLAND REDS AND THE EFFECT OF FEEDING SKIM MILK ON THE CONSTANTS OF THEIR GROWTH CURVES. Jour. Agr. Research 36: 515–540, illus. 1928.

TABLE 2.—*The probable error of the mean weights per chick for each of the four lots together with the standard deviation and coefficient of variation of each mean weight*

Age (in weeks)	Statistical constants	Lot 1 (females)	Lot 2 (females)	Lot 3 (males)	Lot 4 (males)
0	P. E. of mean ..... gm.	± 0.24	± 0.24	± 0.22	± 0.23
	σ ..... gm.	2.02± .17	2.32± .17	2.15± .16	2.16± .16
	C. of V ..... per cent.	5.85± .48	6.78± .50	6.15± .45	6.21± .47
2	P. E. of mean ..... gm.	± 1.88	± 1.32	± 1.56	± 1.50
	σ ..... gm.	16.00± 1.33	12.56± .93	15.21± 1.11	14.06± 1.06
	C. of V ..... per cent.	17.62± 1.51	15.75± 1.20	19.72± 1.49	17.09± 1.33
4	P. E. of mean ..... gm.	± 4.20	± 3.52	± 4.22	± 4.61
	σ ..... gm.	35.26± 2.97	33.44± 2.49	41.04± 2.98	42.73± 3.26
	C. of V ..... per cent.	16.38± 1.42	16.88± 1.31	22.13± 1.69	18.94± 1.50
6	P. E. of mean ..... gm.	± 6.71	± 7.18	± 9.75	± 8.20
	σ ..... gm.	56.25± 4.74	68.19± 5.08	94.77± 6.89	75.92± 5.80
	C. of V ..... per cent.	14.52± 1.25	17.28± 1.33	25.84± 2.00	19.01± 1.50
8	P. E. of mean ..... gm.	± 9.26	± 9.04	±15.07	±12.39
	σ ..... gm.	77.68± 6.55	85.86± 6.39	143.05±10.65	114.70± 8.76
	C. of V ..... per cent.	12.57± 1.08	14.73± 1.12	23.52± 1.85	17.47± 1.37
10	P. E. of mean ..... gm.	±12.57	±11.22	±19.43	±15.74
	σ ..... gm.	105.39± 8.88	106.55± 7.94	184.47±13.74	145.74±11.13
	C. of V ..... per cent.	12.61± 1.08	13.82± 1.05	23.86± 1.88	15.77± 1.23
12	P. E. of mean ..... gm.	±14.89	±11.56	±19.54	±17.54
	σ ..... gm.	122.91±10.53	109.73± 8.17	183.26±13.82	160.31±12.40
	C. of V ..... per cent.	12.47± 1.08	11.71± .88	16.79± 1.30	13.08± 1.03
14	P. E. of mean ..... gm.	±17.61	±12.43	±24.84	±20.03
	σ ..... gm.	145.36±12.45	118.03± 8.79	232.93±17.57	183.03±14.16
	C. of V ..... per cent.	10.89± .94	9.54± .71	15.47± 1.19	11.23± .88
16	P. E. of mean ..... gm.	±19.79	±13.87	±25.24	±19.55
	σ ..... gm.	163.37±13.99	131.67± 9.81	233.64±17.84	178.71±13.83
	C. of V ..... per cent.	10.60± .92	8.89± .66	13.18± 1.03	9.24± .71
18	P. E. of mean ..... gm.	±20.64	±13.91	±25.78	±20.76
	σ ..... gm.	170.38±14.59	132.10± 9.84	238.68±18.23	189.72±14.68
	C. of V ..... per cent.	10.31± .89	8.39± .62	11.62± .90	8.79± .68
20	P. E. of mean ..... gm.	±21.68	±19.59	±27.64	±20.98
	σ ..... gm.	178.96±15.33	186.00±13.85	255.90±19.54	191.76±14.84
	C. of V ..... per cent.	9.62± .82	9.27± .61	10.72± .83	7.87± .61
22	P. E. of mean ..... gm.	±26.48	±23.06	±30.70	±26.82
	σ ..... gm.	218.54±18.72	218.88±16.30	284.16±21.70	241.83±18.96
	C. of V ..... per cent.	10.78± .93	10.88± .82	10.38± .80	8.81± .69
24	P. E. of mean ..... gm.	±32.59	±26.24	±31.90	±28.65
	σ ..... gm.	269.01±23.04	249.14±18.56	295.31±22.55	258.35±20.26
	C. of V ..... per cent.	12.16± 1.06	11.47± .87	10.16± .78	8.83± .69

Table 3 shows the feed consumption in grams per chick for each two-week period for each of the four lots.

TABLE 3.—*Average feed consumption per chick for each two-week period*

Age (in weeks)	Average feed consumption per chick				Age (in weeks)	Average feed consumption per chick			
	Lot 1 (females)	Lot 2 (females)	Lot 3 (males)	Lot 4 (males)		Lot 1 (females)	Lot 2 (females)	Lot 3 (males)	Lot 4 (males)
	Grams	*Grams*	*Grams*	*Grams*		*Grams*	*Grams*	*Grams*	*Grams*
2	127.14	132.76	111.82	136.08	16	1,251.04	1,063.18	1,284.02	1,309.45
4	271.16	246.16	236.29	257.39	18	1,316.88	1,231.34	1,639.91	1,647.26
6	511.28	472.40	489.99	536.40	20	1,534.90	1,305.89	1,520.70	1,715.89
8	688.19	677.62	788.26	814.14	22	1,559.77	1,416.09	1,721.33	1,745.11
10	846.94	824.76	863.49	1,116.54	24	1,613.18	1,261.21	1,625.37	1,764.11
12	1,176.41	1,045.48	1,219.03	1,287.96					
14	1,256.89	1,080.88	1,322.22	1,406.14	Total	12,153.78	10,657.77	12,822.43	13,736.47

APPLICATION OF THE LAW OF DIMINISHING INCREMENT

In order to illustrate the law of diminishing increment graphically the observed average weights of the birds in lot 1 have been plotted as ordinates and the values of the cumulative feed consumption per bird have been plotted as abscissas in Figure 1. The smooth curve passing through the plotted points is the curve of diminishing increment fitted to the last eight of these points. It is readily apparent from the graph that for each successive 1,000 gm. of feed consumed there is a proportionately smaller increase in live weight.

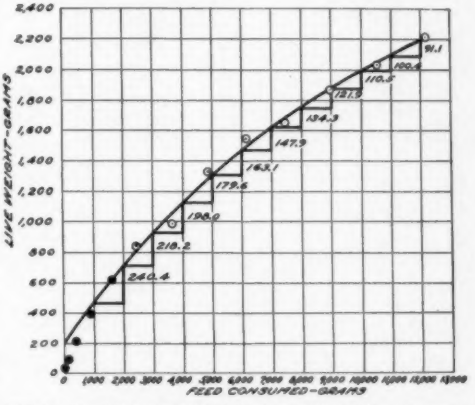

FIG. 1.—Graphic illustration of the law of diminishing increment in chickens; based on the data for lot 1

The number of grams in live weight is shown for each 1,000 gm. of feed consumed, and by calculation it is found that for each successive 1,000 gm. of feed consumed the increase in live weight is approximately 90.7 per cent of the preceding increase in live weight. From the graph one may also read the number of grams of feed required to produce any given average live weight, above 600 gm., of the birds in lot 1.

Table 4 shows for each of the four lots, (1) the average feed consumption per bird per two-week period; (2) the average total feed consumption per bird; (3) the observed average live weight of the birds; and (4) the calculated average live weight of the birds.

TABLE 4.—*Relation between feed consumption and live weight of chickens in lots 1 to 4*

LOT 1 (FEMALES)

Age (in weeks)	Number of birds	Average feed consumption		Average weight per bird		Difference*
		Per period	Total	Observed	Calculated*	
		Grams	*Grams*	*Grams*	*Grams*	*Grams*
0	33			34.5		
2	32	127.14	127.1	90.8		
4	32	271.16	398.3	215.3		
6	32	511.28	909.6	387.3		
8	32	688.19	1,597.8	618.1		
10	32	846.94	2,444.7	835.9	814.1	+21.8
12	31	1,176.41	3,621.1	985.5	1,057.8	−72.3
14	31	1,256.89	4,878.0	1,334.6	1,289.2	+45.4
16	31	1,251.04	6,129.1	1,541.3	1,493.2	+48.1
18	31	1,316.88	7,445.9	1,651.8	1,682.7	−30.9
20	31	1,534.90	8,980.8	1,860.2	1,875.1	−14.9
22	31	1,559.77	10,540.6	2,026.9	2,043.4	−16.5
24	31	1,613.18	12,153.8	2,211.5	2,192.6	+18.9
Root-mean-square deviation						±38.5

* In fitting the curve of diminishing increment only the data obtained after the eighth week were used

TABLE 4.—*Relation between feed consumption and live weight of chickens in lots 1 to 4*—Continued

LOT 2 (FEMALES)

Age (in weeks)	Number of birds	Average feed consumption		Average weight per bird		Difference
		Per period	Total	Observed	Calculated	
		Grams	*Grams*	*Grams*	*Grams*	*Grams*
0	41			34.2		
2	41	132.76	132.8	79.8		
4	41	246.16	378.9	198.1		
6	41	472.40	851.3	355.8		
8	41	677.62	1,528.9	582.7		
10	41	824.76	2,353.7	770.9	756.8	+14.1
12	41	1,045.48	3,399.2	937.0	990.4	−53.4
14	41	1,080.88	4,480.1	1,236.6	1,211.9	+24.7
16	41	1,063.18	5,543.2	1,480.4	1,411.4	+69.0
18	41	1,231.34	6,774.6	1,574.2	1,621.8	−47.6
20	41	1,205.89	7,980.5	1,785.4	1,808.3	−22.9
22	41	1,416.09	9,396.6	2,012.1	2,005.1	+7.0
24	41	1,261.21	10,657.8	2,171.0	2,162.2	+8.8
Root-mean-square deviation						±37.6

LOT 3 (MALES)

0	43			35.0		
2	43	111.82	111.8	77.1		
4	43	236.29	348.1	185.4		
6	43	489.99	838.1	366.8		
8	41	788.26	1,626.3	608.1		
10	41	863.49	2,489.8	773.2	773.2	
12	40	1,219.03	3,708.9	1,091.1	1,116.9	−25.8
14	40	1,322.22	5,031.1	1,505.9	1,457.6	+48.3
16	39	1,284.02	6,315.1	1,773.0	1,759.2	+13.8
18	39	1,639.91	7,955.0	2,046.0	2,106.3	−60.3
20	39	1,520.70	9,475.7	2,386.8	2,394.0	−7.2
22	39	1,721.33	11,197.0	2,736.4	2,684.3	+52.1
24	39	1,625.37	12,822.4	2,906.3	2,927.6	−21.3
Root-mean-square deviation						±35.4

LOT 4 (MALES)

0	40			34.8		
2	40	136.08	136.1	82.3		
4	39	257.39	393.5	225.6		
6	39	536.40	929.9	399.4		
8	39	814.14	1,744.0	656.6		
10	39	1,116.54	2,860.5	924.2	915.9	+8.3
12	38	1,287.96	4,148.5	1,225.7	1,264.9	−39.2
14	38	1,406.14	5,554.6	1,629.7	1,603.6	+26.1
16	38	1,309.45	6,864.1	1,933.0	1,883.7	+49.3
18	38	1,647.26	8,511.3	2,157.4	2,193.6	−36.2
20	38	1,715.89	10,227.2	2,435.5	2,472.2	−36.7
22	37	1,745.11	11,972.3	2,744.9	2,715.9	+29.0
24	37	1,764.11	13,736.5	2,926.9	2,927.2	−.3
Root-mean-square deviation						±32.0

Table 5 shows, for each of the four lots, the expected live weight for each 1,000 gm. of feed consumed. The values in the sixth column of Table 4 and for each of the lots in Table 5 were calculated by means of the formula of the curve of diminishing increment fitted to the data obtained after the eighth week.

TABLE 5.—*The calculated average weight of the birds in each lot per 1,000 gm. cumulative feed consumption per bird*

Average total feed consumption per bird (in grams)	Calculated average weight of the birds				Average total feed consumption per bird (in grams)	Calculated average weight of the birds			
	Lot 1	Lot 2	Lot 3	Lot 4		Lot 1	Lot 2	Lot 3	Lot 4
	Grams	*Grams*	*Grams*	*Grams*		*Grams*	*Grams*	*Grams*	*Grams*
1,000	474.0	423.1	310.6	337.1	8,000	1,755.5	1,811.2	2,115.3	2,102.1
2,000	714.4	673.1	626.5	659.8	9,000	1,877.4	1,952.3	2,307.3	2,277.2
3,000	932.6	903.6	920.7	955.6	10,000	1,987.9	2,082.3	2,486.2	2,437.7
4,000	1,130.6	1,115.9	1,194.5	1,226.6	11,000	2,088.3	2,202.1	2,652.8	2,584.7
5,000	1,310.2	1,311.6	1,449.9	1,474.9	12,000	2,179.4	2,312.5	2,807.9	2,719.4
6,000	1,473.3	1,491.9	1,687.7	1,702.5	13,000	2,262.0	2,414.3	2,952.6	2,842.9
7,000	1,621.2	1,658.1	1,909.1	1,911.0	14,000	2,337.0	2,508.0	3,087.1	2,956.0

The method used in fitting the curve of diminishing increment to the writers' experimental data is essentially different from the methods described by Spillman [6] and for that reason is given in some detail here. No originality is claimed for this method; it is a combination of methods which have been used by others. The general formula for the curve of diminishing increment, as written by Spillman, is

$$Y = M - AR^x \quad (1)$$

in which

Y = live weight for any given amount, x, of feed consumed.
M = theoretical maximum live weight attainable,
A = the gain in live weight, above the *theoretical* initial weight, of which the birds are capable if their growth continues to obey the same law,
R = ratio between gains in live weight for successive units of feed, and
x = the number of units of feed consumed.

The first step is to transform the equation into its linear form, which is:

$$\log A + x \log R - \log (M - Y) = 0 \quad (2)$$

If one now selects a value for M and plots log (M − Y) against X, an approximately straight line results, if the selected value of M is sufficiently close to its true value. By trying several different values for M, the true value may thus be approximated. Having obtained an approximate value of M, the next step is to substitute it in equation (2) and then determine the values of A and R by the method of least squares. (Figs. 2 to 5.)

Having approximate values for M, A, and R, one is now ready to "adjust" them by the usual method.

The authors have found that the computations incident to making these adjustments are very much simplified if equation (1) is rewritten as follows:

$$Y = M - A\epsilon^{-kx} \quad (3)$$

[6] SPILLMAN, W. J., and LANG, E. Op. cit.

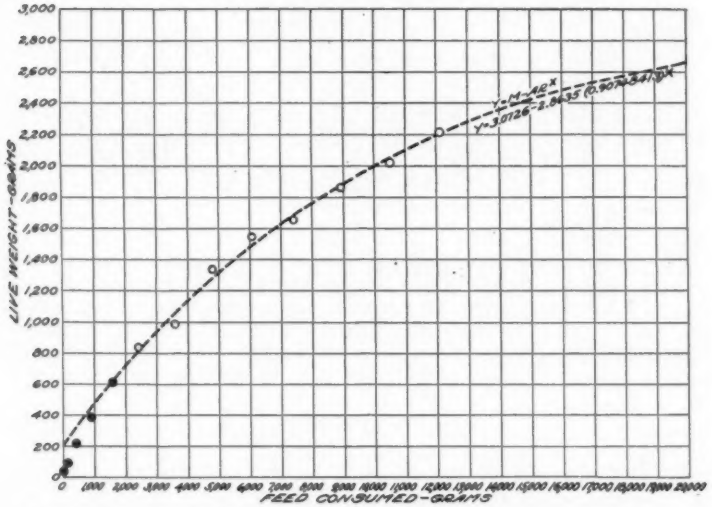

FIG. 2.—Relation between weight of birds in lot 1 (females) and average amount of feed consumed

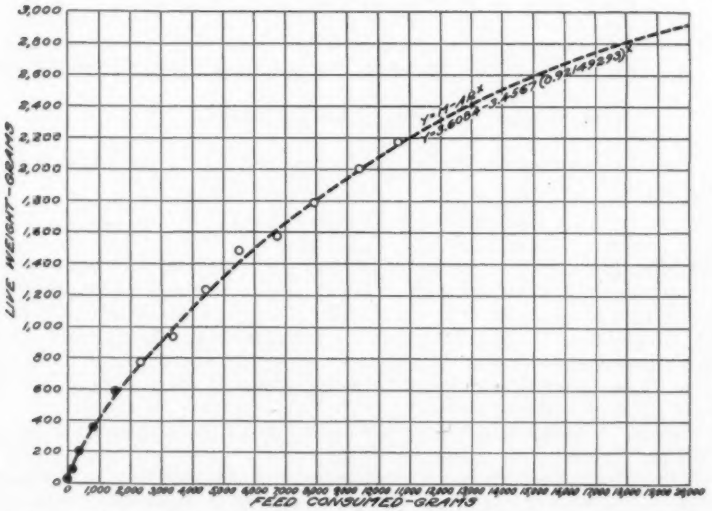

FIG. 3.—Relation between weight of birds in lot 2 (females) and average amount of feed consumed

in which ϵ^{-k} replaces R of equation (1). The "adjustment" equation for this form of the equation of the curve of diminishing increment is:

$$\frac{\delta f}{\delta M_0}\mu+\frac{\delta f}{\delta A_0}\alpha+\frac{\delta f}{\delta k_0}\kappa=Y-Y_0 \quad (4)$$

in which

$$\frac{\delta f}{\delta M_0}=1,$$

$$\frac{\delta f}{\delta A_0}=\epsilon^{-k_0 x},$$

$$\frac{\delta f}{\delta k_0}=xA_0\epsilon^{-k_0 x}.$$

μ, α, and κ are corrections to be made to M_0, A_5 and k_0, respectively, which are the approximate values of M, A and R previously obtained, and Y and Y_0 are, respectively, the observed and calculated values of Y.

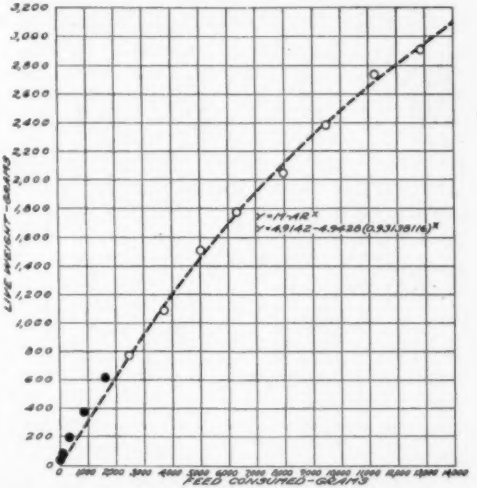

FIG. 4.—Relation between weight of birds in lot 3 (males) and average amount of feed consumed

By readjusting the corrected values of M, A, and k until the corrections become negligible (two to three adjustments were found to be necessary), one finally obtains with considerable accuracy the most probable values of these constants. It is now a simple matter to calculate the most probable value of R from the relationship between R and k, that is, $R=\epsilon^{-k}$.

The above method was applied to each of the four sets of data, and the values of the constants shown in Table 6 were obtained.

TABLE 6.—*Constants of the curve of diminishing increment* [a]

Lot	M	A	R	k [b]	S [c]
	Kilograms	*Kilograms*			*Kilogram*
1, females	3.0726	2.6635	0.907484	0.0970791	0.0385
2, females	3.6084	3.4567	.921493	.0817604	.0376
3, males	4.9142	4.9428	.931381	.0710867	.0354
4, males	4.1954	4.2105	.916351	.0873561	.0329

[a] The unit of live weight, as well as the unit of feed weight used in determining these constants, was the kilogram; the same unit should be used in making any calculations involving these constants.
[b] See equation (3).
[c] S is the root-mean-square deviation of the observed values of Y from the calculated values of Y.

The differences between the observed and calculated values of the average live weights for each of the four lots (column 7, of Table 4), were found to be relatively small. It was also found that the root-mean-square deviation of the calculated from the observed live weights was of the same order of magnitude for each of the four lots.

It is rather difficult to explain the difference between the values of M for lots 1 and 2, as well as for lots 3 and 4, since these were duplicate lots which received the same treatment throughout. The same may be said of the values of A. Tables 1 and 4 show, however, that these duplicate lots were not strictly comparable [7] at the tenth week; and it was to the data obtained from the tenth to the twenty-fourth week that the curve of diminishing increment was fitted.

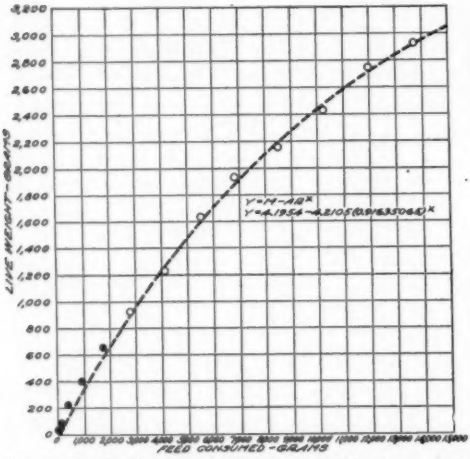

FIG. 5.—Relation between weight of birds in lot 4 (males) and average amount of feed consumed

In the case of the constant R, the values obtained for each of the four lots are approximately the same; the average value of R is very nearly 0.92.

CONCLUSION

It seems, therefore, that, so far as the writers' results are concerned, M and A can be considered only as empirical constants and should not be thought of as representing, respectively, the maximum live weight *actually* to be attained and the *actual* gain necessary to reach that weight. In any case, the values of R indicate that, for the feed interval to which the curve of diminishing increment was fitted, the ratio between gains in live weight for successive kilogram units of feed is approximately 0.92. In view of the agreement between the observed and calculated live weights, the writers are led to the conclusion that the relationship between feed consumption and growth of the domestic fowl is expressible by the law of diminishing increment, at least for the feed interval studied.

[7] The most probable reason that these lots were not strictly comparable at the tenth week is that different amounts of feed had been consumed by the chicks in each of the pens. The reason for this difference in feed consumption is not clear. However, since some of the feed was fed according to the judgment of the attendant as to how much the birds would eat, and since in two of the pens the number of birds was smaller than in the other two pens, it is probable that more feed, per chick, was received by the birds in the pens containing fewer birds than in those containing the larger number. This suggestion as to the cause of the observed differences agrees with the facts.

THE ACCURACY OF CATTLE WEIGHTS[1]

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INTRODUCTION

It has been known for a long time that weights of cattle are subject to considerable fluctuation from day to day. Such fluctuations are of decided importance to research workers who are conducting experiments involving gains or losses in the weight of cattle. In a general way two classes of fluctuations may be distinguished: The first consists of sudden, rather large changes in the average weight of a group in which all, or nearly all, of the cattle gain (or lose) together, although not always to the same extent. Such changes may properly be called "fills" or "shrinks" in accordance with market usage of these terms, and the causes of such changes are usually readily recognizable in some environmental circumstances, such as a sudden drop in temperature, shipment to market, turning on pasture, etc. The second class consists of daily weight changes (either gains or losses) which individual animals may make, and which are independent of the daily trend of the average weight of their group. Both classes of weight fluctuations are encountered in feeding experiments. As an illustration of extreme and minimal "shrinks" and "fills" in weight among groups of experimental cattle may be cited the weights obtained on some dairy cows in a feeding experiment at College Station, Tex., in 1927. Twenty-four cows were weighed for three successive days in February, and 19 of the same cows, together with 10 others, were weighed for three successive days in April. The average weights were as follows:

	Feb. 16–18	Apr. 13–15
First day	856 pounds	824 pounds
Second day	849 pounds	822 pounds
Third day	813 pounds	821 pounds

The variation found in the February weights in this case was the most extreme in the writers' observation of dairy cows, yet in several other instances the range between the largest and smallest of these average weights for three successive days has exceeded 20 pounds. The weather records show clearly the cause of the heavy shrink from the second to the third weight in February, since in the 23 hours preceding the third day's weight the temperature dropped 52° F. and during part of that time a light mist fell. The cows were miserable in the freezing temperature, to which they were unaccustomed, and

[1] Received for publication Dec. 16, 1927; issued May, 1928.

[2] The statistical methods used in this study were selected and the calculations were made by the senior author. Indebtedness is hereby acknowledged to the following men for their part in collecting the original data and for helpful suggestions in interpreting these data: J. H. Shepperd, North Dakota Agricultural Experiment Station; F. E. Keating, of the Big Spring Station, Bureau of Plant Industry, U. S. Department of Agriculture; J. M. Jones, Fred Hale, R. E. Dickson, J. H. Jones, and W. E. Flint, of the Texas Agricultural Experiment Station; and E. W. Sheets, R. H. Tuckwiller, A. T. Semple, Bradford Knapp, jr., and E. W. McComas, of the Bureau of Animal Industry, U. S. Department of Agriculture.

naturally drank very little water. Possibly such variations may ordinarily be greater in mature dairy cows than in beef steers, but that large daily fluctuations do occasionally occur even among large groups of beef steers may be illustrated by the following average weights for a group of 72 North Dakota steers in the Mandan grazing trials for 1923. The small first weight is doubtless due to the fact that the steers were weighed directly from the cars after a shipment (from Mandan to Fargo) which caused them to be without feed and water for over 24 hours.

Date weighed:	Average weight in pounds	Date weighed:	Average weight in pounds
October 30	823	November 3	886
October 31	876	November 5	894
November 1	885	November 6	902
November 2	887	November 7	900

THE USE OF CONTROL LOTS

Research workers generally have sought to eliminate the experimental errors arising from such group fluctuations in weight by using control or check lots and studying not the absolute gain or loss of the experimental lot in pounds but the difference between such gain or loss and the gain or loss made by the control lot which is weighed on the same day and is given as nearly as possible the same treatment as the experimental lot except for the one variable which is the object of study. Undoubtedly this procedure has eliminated much of the experimental error which would be involved in using the actual weights without comparison with the weights of a control lot. However, because individual animals do not vary in weight exactly as their group average does, not all the error is thus eliminated. For example, in the weights given above for the North Dakota steers between November 6 and November 7 steer No. 85 gained 20 pounds, while steer No. 88 lost 15 pounds, steer No. 90 lost 10 pounds, and steer No. 91 gained 10 pounds. As long as such fluctuations independent of the daily trend of the group occur, there is always the possibility that the control lot will by "chance" contain a larger number of plus or of minus fluctuations than the experimental lot, and this may lead to more or less serious errors in interpreting the data.

This leads to the precise question which is the subject of study in this paper: How large are the experimental errors which can not thus be eliminated by the use of control lots in carefully conducted experiments, and are there any recognizable circumstances, such as the age, size or gentleness of the cattle, type of ration being fed, and weather conditions, which cause this experimental error to be larger or smaller than normal and which can be used in estimating the size of the error even before the weights are taken, if necessary.

WEIGHING FOR THREE OR MORE DAYS

Most research workers have sought to eliminate this experimental error by weighing the cattle for three successive days and using the average of the three weights thus obtained as the correct weight instead of using a single day's weight. Doubtless the use of three-day average weights has helped to eliminate much of the experimental error caused by the "chance" fluctuations of individual

weights. Still, no one who has studied the subject would maintain that all of the experimental error has thus been eliminated. One need only weigh cattle for four successive days to find out that the fourth day's weights do not correspond exactly to the average weight for the first three days, nor does the four-day average correspond exactly to the three-day average. Armsby (*1*)[3] long ago pointed out that even a 10-day average weight did not entirely eliminate the influence of daily fluctuations in weight. Moreover, it sometimes happens that there are serious practical objections to weighing for three successive days. For example, it frequently happens in the case of grazing experiments that the scales are located at a considerable distance from some of the pastures, and the investigator may well wonder whether the unusual amount of excitement and exercise to which his cattle are subjected in being rounded up and driven in to the scales on the two additional days does not introduce errors greater than are eliminated by weighing for three days instead of one. It is therefore very desirable for the investigator to know just what increase in accuracy is gained by weighing for three days instead of one.

SCOPE OF THIS STUDY

The results of a preliminary study of this subject were presented at the annual meeting of the American Society of Animal Production at Chicago in November, 1926, and an abstract of the report there made appears in the proceedings of the meeting (*7*). The interest there expressed was largely responsible for a determination to enlarge the study to its present scope and to include weights taken under such widely different climatic conditions as prevail in two different regions in North Dakota (Mandan and Fargo), one in West Virginia (Greenbrier County), and four in Texas (Big Spring, College Station, Kingsville, and Spur). The data include weights of calves, yearlings, and 2-year-olds, thin and fat, in feed lots; yearlings, 2-year-olds, and 3-year-olds on pasture in the spring and in the fall, and dairy cows. Cattle of Hereford, Shorthorn, Aberdeen-Angus, Jersey, and Brahman breeding are included. Altogether, 9,897 individual weights were taken on more than 900 different animals. All animals were weighed at least on 3 consecutive days and some were weighed on as many as 11 consecutive days. Statistically, the total amount of data is equivalent to that which would be obtained by weighing 3,360 animals individually on 3 consecutive days, 672 animals on 11 consecutive days, or 1 animal, whose weight was not showing any consistent increase or decrease under absolutely uniform weather and other environmental conditions, on 6,721 consecutive days.

SPECIAL PECULIARITIES OF THE DATA STUDIED

The weights were not all taken with the same degree of precision. Thus most of the Kingsville weights and many of the Fargo weights and some of the Spur weights were taken to the nearest pound; the dairy weights, most of the Spur weights, and some of the Big Spring weights were taken to the nearest 2 pounds, while the West Virginia weights, Mandan weights, and most of the Big Spring weights were taken to the nearest 5 pounds.

[3] Reference is made by number (italic) to "Literature cited," p. 579.

Some of the weights were taken on cattle which were rather wild and excitable (as the initial Kingsville weights), and other weights were taken on cattle which had become rather gentle (as the final weights in the feeding experiments), or on cattle which had been thoroughly gentle for a long time (as the dairy weights).

Some of the cattle were getting nothing to eat but pasture; others were getting both pasture and concentrates; many others were getting concentrates, dry roughage, and large amounts of silage; others were getting only concentrates and dry roughage and were confined at all times to the feed lot; and still others were getting dry roughage while on pasture.

Some of these peculiarities doubtless affect the accuracy of the individual weights, and these effects will be considered later.

STATISTICAL METHODS USED IN THE PRELIMINARY STUDY

In the preliminary report made at the 1926 meeting of the American Society of Animal Production, multiple correlation methods were used to calculate the standard error of estimating the average weight for an infinite number of days from a single day's weight or from the average of n days' weights. This calculation involved the assumption that the average correlation found between the weight of the same animals on the different days when they actually were weighed was the average correlation which would be found between weights of these same animals for any two different days if weighed for an infinite number of days. It also involved the assumption that the average of the three or more standard deviations actually found for the weights on the three or more days when weights were taken was the standard deviation of the weights of that particular group of cattle on any particular day. The error introduced by these assumptions was very slight, but was, nevertheless, undesirable. Since the printing of that report the senior author's attention was called to the fact that, by the use of intraclass correlations, difficulties in averaging correlations and standard deviations could be avoided, and thus both the above assumptions could be reduced to the assumption that the data actually studied were a random sample of other weights which might be taken on the same group of cattle, an assumption which is inherent in almost all statistical studies.

The method used in the preliminary report also differed in two other respects from the method used in the present study. In the first place, the ordinary biometrical formula for the standard deviation $\left(\sqrt{\frac{\Sigma d^2}{n}}\right)$ was used in the former, while the analysis of variance which was employed in the present study necessarily makes use of what is known as Bessel's formula $\left(\sqrt{\frac{\Sigma d^2}{n-1}}\right)$ or some modification of it to correspond to the number of "degrees of freedom" [4] in the data. In the second place, the standard errors given in the preliminary report were the standard errors of estimating the average of an infinite number of weights (as the dependent variable) from a single weight or from an average of two or more days' weights (as the

[4] See Fisher's Statistical Methods for Research Workers, p. 53 and 54, for discussion of which formula is most appropriate to use, and Chapters VII and VIII for further explanation of the idea of degrees of freedom (3).

independent variable). Now in actual practice investigators do not allow for the idea of regression in their estimate of the average of an infinite number of weights. Instead they say, "The average of the three days' weights was taken as the true weight," or some similar statement. The standard error of such estimates which ignore regression is the standard error of estimating (by the correct regression equation) a single weight or a two-day or three-day average weight (as the dependent variable) from the average of an infinite number of weights (as the independent variable). Both of these ideas can be incorporated in the multiple correlation method, and when that is done the results of that method and the "analysis of variance" method used in this study become identical, as of course they should if both methods are correct in principle.[5]

The difference between the figures as printed in the original report and as calculated in this study is very small and is almost entirely due to the use of Bessel's formula in this study instead of the usual biometrical formula which ignores the loss of degrees of freedom. Table 1 shows the standard errors as originally printed and as calculated by the analysis of variance.

TABLE 1.—*The experimental error in single weights of cattle as printed in the preliminary report and as calculated by the analysis of variance*

Experiment studied	Standard error of a single weight	
	As printed in the preliminary report	As calculated by the analysis of variance
	Pounds	*Pounds*
First Kingsville, initial	[a] 6.08	[a] 6.66
First Kingsville, final	[a] 6.35	7.41
First Big Spring, initial	[b] 7.06	7.29
First Big Spring, final	[a] 6.06	6.61
Second Big Spring, initial	5.67	5.83
Second Big Spring, final	[a] 7.94	8.56
Third Big Spring, initial	5.04	5.42
Third Big Spring, final	[a] 4.63	5.34

[a] These figures were so calculated that the error is figured from the daily trend within each lot as a base instead of from the daily trend of the average weight of all lots combined. This has the effect of an assumption that differences in the daily trends of the weights of two lots of steers in the same experient on the same weighing days are due to external differences in the ration or treatment and are not due to plus or minus fluctuations which are by chance greater in one lot than in the other. Either view might be correct according to the objects and conditions of each experiment.

[b] Through error in calculation this figure was printed as 7.49 in the preliminary report.

STATISTICAL METHODS USED IN THE PRESENT STUDY

Fisher's method (*3*) of analyzing variance (as explained in detail in Chapters VII and VIII of Statistical Methods for Research Workers) gives a "remaining variance" or "experimental error" which is identical with the standard error of estimating by multiple correlation methods a single day's weight from an average of an infinite number of weights, provided Bessel's formula is used for calculating the standard deviations. Fisher's method involves the idea of "degrees of freedom" which is involved in Bessel's formula for the standard deviation $\left(\sqrt{\frac{\Sigma d^2}{n-1}}\right)$, but is not involved in the for-

[5] The writers are especially indebted to Sewall Wright, of the University of Chicago, for his kindness in checking the statistical formulas derived for use in the multiple correlation method.

mula ordinarily used by biometricians $\left(\sqrt{\frac{\Sigma d^2}{n}}\right)$. Statistical workers are rather generally agreed that the latter formula is the correct one where the standard deviation is to be applied to the sample from which it was calculated, as in calculating correlations or in fitting curves, but that the idea of "degrees of freedom" should be taken into consideration if the calculated standard deviation is to be regarded as an estimate of the true standard deviation existing in a very large population of which the actual data used in the calculation are only a random sample. The difference between the standard deviations as calculated by the two formulas is very small if n is at all large, being less than 4 per cent if n is as large as 12 and less than 1 per cent if n is as large as 50. Such difference as does exist causes the experimental error in cattle weights as figured by Fisher's method to be very slightly larger than the original figures given in the preliminary report for the errors in the one-day weights of cattle.

Briefly, the method of the analysis of variance which is used in this study consists of separating the total variance, Σd^2, of all the individual weights into three portions: First, a portion due to the fact that the daily average weight is not identical on all three (or more) days of weighing; second, a portion due to the fact that the average weights of the different cattle for the whole period weighed are not identical; and finally, a remaining portion which is not due to either of these facts and which may properly be considered as experimental error. It is this remaining portion which is the object of study in this paper. Perhaps this remaining portion can be visualized better if we imagine a group of steers each of which is weighed for three (or more) days and all of which have identical average weights for the period of three days, and if we further imagine that weather and treatment were so uniform and equable during the days of the weighing that the average weights of the entire group for the different days were identical. Then in such a group of individual weights the standard deviation (with appropriate corrections for the number of degrees of freedom sacrificed in meeting the conditions imposed) would be the experimental error as found by the analysis of variance (or the standard error of estimating a single weight from the average of an infinite number of daily weights as found by the multiple correlation method). This amount of variation could be large or small, as illustrated by the hypothetical figures given in Table 2, in which the first two animals show a small degree of variation from day to day, while the last two show a large degree of variation.

TABLE 2.—*Hypothetical weights showing day-to-day variation possible without variation in average weights*

Steer No.	Hypothetical weights for—			Total
	First day	Second day	Third day	
	Pounds	*Pounds*	*Pounds*	*Pounds*
1	910	912	908	2,730
2	912	912	906	2,730
3	890	930	910	2,730
4	928	886	916	2,730
Total	3,640	3,640	3,640	10,920

To illustrate the method of calculation, in Table 3 are reproduced the actual data for the lot 3 West Virginia steers in 1925, and in Table 4 is given a summary of the analysis of the variance in the actual data presented in Table 3. The formulas and principles for obtaining the figures in Table 4 may be found in Fisher's book (*3*). Calculations similar to those in Table 4 were made for every group of cattle studied, and the standard deviations due to experimental error were then subjected to further study.

TABLE 3.—*Weights of lot 3, West Virginia 3-year-old steers, 1925*

Steer No.	Weight of steers on—			Total	Steer No.	Weight of steers on—			Total
	Apr. 21	Apr. 22	Apr. 23			Apr. 21	Apr. 22	Apr. 23	
	Pounds	*Pounds*	*Pounds*	*Pounds*		*Pounds*	*Pounds*	*Pounds*	*Pounds*
3	920	925	910	2,755	51	960	965	975	2,900
6	830	820	815	2,465	54	940	975	960	2,875
9	775	770	770	2,315	57	1,050	1,040	1,040	3,130
12	1,090	1,105	1,085	3,280	60	1,010	1,015	1,000	3,025
15	1,015	1,000	1,000	3,015	61	890	875	895	2,660
18	850	860	855	2,565	66	945	945	950	2,840
19	870	850	845	2,565	69	885	880	870	2,635
24	905	905	905	2,715	72	930	920	910	2,760
27	845	845	845	2,535	75	960	965	955	2,880
30	960	950	940	2,850	78	1,035	1,040	1,050	3,125
33	1,065	1,050	1,065	3,180	81	1,070	1,070	1,060	3,200
36	1,010	1,020	1,025	3,055	84	880	875	870	2,625
39	965	975	970	2,910	87	1,080	1,060	1,055	3,195
42	1,085	1,075	1,075	3,235	90	1,020	1,010	1,010	3,040
45	960	960	935	2,855					
48	940	940	920	2,800	Total	28,740	28,685	28,560	85,985

TABLE 4.—*Summary of the analysis of the variance in lot 3, West Virginia steers, for April, 1925*

Variance due to—	Degrees of freedom	Sum of squares	Mean square	Standard deviation
All causes	89	617,911.388	6,942.82	83.32
Daily trend	2	567.222	283.61	3.07
Remainder	87	617,344.166	7,095.91	84.24
Difference between animals	29	613,461.388	21,153.84	83.97
Final remainder or experimental error	58	3,882.778	66.94	8.18

Table 4 is fairly representative of all such tables. In every case differences between animals were by far the most important causes of variation in the data. In nearly every instance the daily trend was a comparatively unimportant source of variation, although in a few cases it was important, and in the first Kingsville initial weights and in the February dairy cattle weights its standard deviation actually exceeded the experimental error.

RESULTS OF THIS STUDY

A summary of the experimental error actually found in the different groups of cattle is presented in Table 5, along with other pertinent facts about the age, weights, ration, etc.

TABLE 5.—*Average weight of cattle, together with precision of weights, standard deviation of average weights, degrees of freedom, and experimental error found in the different groups of weights*

Group of weights studied	Number of cattle	Approximate age	Number of days weighed	Average weight (pounds)	Precision of weights (pounds)	Standard deviation of average weights	Degrees of freedom	Experimental error (pounds)	Notes [a]
First Kingsville, initial	100	9 to 11 months	3 and 4	529	1	77	216	6.7	A.
First Kingsville, final	67	15 to 17 months	3	795	1	57	132	7.4	
Third Kingsville, initial	99	16 to 22 months	3	628	[b] 5—	73	196	11.9	B.
Third Kingsville, final	98	22 to 28 months	3	895	1	82	194	10.7	
First Big Spring, initial	42	7 to 9 months	3	427	[c] 5 and 2—	39	82	7.3	C.
First Big Spring, final	45	13 to 15 months	3	717	5—	69	88	6.6	
Second Big Spring, initial	45	8 to 10 months	3	506	5—	45	88	5.8	D.
Second Big Spring, final	45	14 to 16 months	3	860	5—	68	88	8.6	
Third Big Spring, initial	45	6 to 8 months	3	308	5—	26	88	5.4	E.
Third Big Spring, final	44	13 to 15 months	3	666	5—	74	86	5.3	
Spur, 1922–23:									
2-year-olds, initial	22	32 months	3	840	2	86	42	14.6	F.
2-year-olds, final	21	36 months	3	1,179	2	134	41	11.2	
Yearlings, initial	26	20 months	3	542	2	67	50	8.3	G.
Yearlings, final	25	24 months	3	876	2	94	48	6.2	
Spur, 1924–25:									
2-year-olds, initial	24	32 months	3	780	2	96	46	11.5	H.
2-year-olds, final	24	36 months	3	1,131	2	131	46	15.2	
Yearlings, initial	20	20 months	3	586	2	81	38	14.9	H.
Yearlings, final	20	24 months	3	857	2	85	38	10.5	
Spur, 1925–26:									
Yearlings, initial	29	21 months	3	632	2	74	56	6.2	I.
Yearlings, final	29	25 months	3	947	2	83	56	7.3	
Calves, initial	32	9 months	3	422	2	46	62	6.1	I.
Calves, final	31	13 months	3	686	2	68	60	7.2	
Spur, 1926–27:									
Calves, initial	36	10 months	3	398	1	71	70	4.8	J.
Calves, final	36	14 months	3	633	1	94	70	5.3	
Swenson, initial	30	14 months	3	582	1	97	58	10.5	K.
Swenson, final	30	18 months	3	795	1	106	58	8.9	
College station dairy cows:									
In rice-bran project, November, 1925	18	2 to 12 years	3	847	2	57	34	10.9	L.
In cottonseed-meal project—									
December, 1926	25	do	3	870	2	101	48	12.1	
January, 1927	24	do	3	836	2	102	46	10.1	
February, 1927	24	do	3	839	2	105	46	13.1	
March, 1927	34	do	3	804	2	105	66	16.0	
April, 1927	29	do	3	822	2	105	56	11.6	
May, 1927	28	do	3	860	2	112	54	13.5	
June, 1927	30	do	3	871	2	110	58	8.2	
July, 1927	26	do	3	848	2	108	50	12.1	

West Virginia:									
Calves, fall, 1922—									
Lot 1	30	6 months	3	388	5	42	58	8.1	M.
Lot 2	30	do	3	382	5	42	58	7.9	
Lot 3	30	do	3	389	5	48	58	6.0	
Yearlings, spring, 1923—									
Lot 1	30	1 year	3	446	5	46	58	7.3	N.
Lot 2	30	do	3	462	5	55	58	6.6	
Lot 3	30	do	3	499	5	51	58	6.7	
Yearlings, fall, 1923—									
Lot 1	30	1½ years	3	576	5	51	58	8.2	O.
Lot 2	30	do	3	575	5	55	58	8.6	
Lot 3	30	do	3	576	5	54	58	9.1	
2-year-olds, spring, 1924—									
Lot 1	30	2 years	3	662	5	64	58	9.4	P.
Lot 2	30	do	3	615	5	60	58	11.5	
Lot 3	30	do	3	655	5	61	58	11.5	
2-year-olds, fall, 1924—									
Lot 1	30	2½ years	3	898	5	75	58	13.7	Q.
Lot 2	30	do	3	899	5	70	58	14.5	
Lot 3	30	do	3	916	5	76	58	16.9	
3-year-olds, spring, 1925—									
Lot 1	30	3 years	3	929	5	89	58	13.2	R.
Lot 2	30	do	3	936	5	90	58	11.3	
Lot 3	30	do	3	955	5	84	58	8.2	
North Dakota steers:									
Mandan grazing trials—									
Oct.–Nov., 1922	64	2½ years	6	913	5	86	315	17.7	S.
May, 1923	93	2 years	3	678	2	100	184	8.1	T.
Oct.–Nov., 1923	72	2½ years	8	882	5	96	497	16.1	U.
Oct.–Nov., 1923	6	do	7	849	5	51	30	16.7	V.
1921 grazing sweet-clover trials—									
May 3–5	6	2 and 3 years	3	1,027	5	190	10	8.1	W
May 9–11	6	do	3	998	5	193	10	13.3	
May 12–18	4	2 years	6	860	1	62	15	12.7	
May 23–28	6	2 and 3 years	6	976	1	151	25	5.9	
June 1–8	6	do	6	1,019	1	152	25	11.2	
July 14–16	6	do	3	1,105	5	146	10	17.1	
Sept. 14–16	6	2½ and 3½ years	3	1,181	5	160	10	8.0	
1922–23 alfalfa hay and corn silage, steers—									
Nov. 27–29	24	2½ years	3	954	5	110	46	9.1	X.
Dec. 27–29	24	do	3	1,024	5	124	46	8.7	
Jan. 26–28	23	do	3	1,105	5	104	44	7.1	
Feb. 15–17	22	3 years	3	1,149	5	113	42	9.8	

[a] The notes referred to in this column will be found immediately following the table.

[b] The minus sign signifies that the degree of precision was slightly greater than the figure indicates. In this case most of the steers were weighed to the nearest 5 pounds, but a few were weighed to the nearest 2 pounds.

[c] These steers were weighed to the nearest 5 pounds on the first day, but most of them were weighed to the nearest 2 pounds or less on the second and third days.

TABLE 5.—*Average weight of cattle, together with precision of weights, standard deviation of average weights, degrees of freedom, and experimental error found in the different groups of weights*—Continued

Group of weights studied	Number of cattle	Approximate age	Number of days weighed	Average weight (pounds)	Precision of weights (pounds)	Standard deviation of average weights	Degrees of freedom	Experimental error (pounds)	Notes
North Dakota steers—Continued.									
1922–23 sweet-clover hay and corn silage, steers—									
Nov. 27–29	24	2½ years	3	961	5	111	46	8.5	Y.
Dec. 27–29	24	do	3	1,023	5	120	46	6.9	
Jan. 26–28	24	do	3	1,088	5	126	46	7.8	
Feb. 15–17	24	3 years	3	1,118	5	130	46	8.6	
Silage experiments, 1921–22—									
Dec. 8–10	20	2½ years	3	904	1	75	38	10.4	Z.
Jan. 7–9	20	do	3	1,015	1	87	38	7.3	
Feb. 6–8	20	do	3	1,036	1	94	38	8.6	
Mar. 8–10	19	do	3	1,081	1	92	36	9.4	
Mar. 18–20	19	3 years	3	1,088	1	89	36	10.1	
Silage experiments, 1922–23—									
Nov. 21–Dec. 1	19	2½ years	11	907	1	31	180	12.4	AA.
Dec. 9–11	19	do	3	956	1	32	36	12.0	
Dec. 29–31	19	do	3	975	1	54	36	12.5	
Jan. 29–31	19	do	3	1,008	1	62	36	8.8	
Feb. 27–Mar. 1	19	do	3	1,026	1	70	36	7.9	
Mar. 29–Apr. 1	19	3 years	4	1,099	1	81	54	9.5	
Silage experiments, 1923–24—									
Dec. 15–22	20	2½ years	8	892	1	50	133	10.1	AB.
Dec. 30–Jan. 1	20	do	3	918	1	51	38	15.1	
Jan. 19–21	20	do	3	957	1	58	38	8.9	
Feb. 18–20	20	do	3	1,002	1	69	38	12.6	
Mar. 19–21	20	do	3	1,057	1	82	38	9.4	
Apr. 18–20	20	do	3	1,112	1	95	38	7.7	
Apr. 27–29	20	3 years	3	1,130	1	99	38	9.1	
Silage experiments, 1924–25—									
Nov. 11–21	20	2½ years	11	960	1	48	190	13.9	AC.
Dec. 19–21	20	do	3	1,055	1	53	38	9.9	
Jan. 17–19	20	do	3	1,107	1	59	38	16.5	
Feb. 16–18	20	do	3	1,160	1	76	38	14.5	
Mar. 19–21	20	do	3	1,209	1	96	38	7.7	
Apr. 8–10	20	3 years	3	1,230	1	102	38	8.9	
Silage experiments, 1925–26—									
Nov. 22–27	20	2½ years	6	930	1	63	95	11.8	AD.
Dec. 5–7	20	do	3	958	1	65	38	9.1	
Dec. 25–27	20	do	3	995	1	65	38	11.7	
Jan. 24–26	20	do	3	1,045	1	75	38	7.9	
Feb. 23–25	20	do	3	1,104	1	82	38	11.6	
Mar. 25–27	20	do	3	1,168	1	94	38	5.5	
Apr. 14–16	20	3 years	3	1,200	1	95	38	9.2	

NOTES

A. These calves had just been separated from their dams on the range. One lot consisted of Herefords, another of Shorthorns, another of crossbred calves out of Hereford cows but by high-grade Brahman bulls; the fourth lot consisted of crossbred calves out of Shorthorn cows but sired by high-grade Brahman bulls. They were fed for 179 days in dry lots on a fattening ration of corn or grain sorghum, cottonseed meal, Rhodes grass hay, and a little sorghum silage.

B. These steers had been grazing on fairly good pasture but without grain for several months before the feeding began. Two lots consisted of Hereford and Shorthorn steers, the third lot consisted of crossbred steers out of Shorthorn cows but sired by high-grade Brahman bulls, and the fourth lot consisted of steers out of cows and by bulls identical in breeding with the third lot of steers. They were fed in dry lots for 175 days on fattening rations of ground grain sorghums or ground corn, cottonseed meal, and Rhodes grass hay or hegari stover.

C. These calves were Herefords which had been weaned about two weeks when the experiment began and had been on preliminary feed in dry lots during that time. They were fed in dry lots for 175 days on fattening rations of ground milo heads, cottonseed meal, and sorgo fodder or cottonseed hulls or sorgo silage and Sudan grass hay. Described in more detail in Bulletin No. 363 of the Texas station (*4*).

D. Same as C, except that the calves had been weaned, dehorned, and branded just three days before the experiment began and the fattening period was only 168 days.

E. Same as C, except that the calves had been weaned only three days before the experiment began and the fattening period was 203 days in length.

F. These steers were Herefords which had been on rather poor pasture all summer and were quite thin. One lot was fed cottonseed meal and hulls. The other lot received ground shelled corn in addition. They were full fed for 120 days in a dry lot. Described in detail in Bulletin 309 of the Texas station (*5*).

G. These steers had been on rather poor pasture all summer and were quite thin. One lot consisted of Herefords and the other lot consisted of crossbred steers out of Hereford dams but sired by high-grade Brahman bulls. They were full fed for 120 days in a dry lot on a ration of ground shelled corn, cottonseed meal, and hulls.

H. These steers had been on good pasture until late in the summer and had been shipped to Spur about two weeks before the initial weights were taken. Half of them of each age were Herefords and the rest were crossbred Hereford-Brahmans, like those described in G. They were full fed for 112 days on a ration of ground milo heads, cottonseed meal, cottonseed hulls, and sorghum hay.

I. These steers had been on good pasture until the middle of October, when they were shipped to Spur. They were fed a moderate grain and sorghum fodder ration in dry lots at Spur for a month before the first weights were taken. There were three lots of each age. One lot consisted of Herefords and one lot of Hereford-Brahman crossbreds. The third lot was sired by a Hereford bull out of Hereford-Brahman crossbred cows. They were full fed in a dry lot for 112 days on a fattening ration of ground milo heads, cottonseed meal, and chopped sorghum fodder.

J. These calves were of the same three kinds of breeding as those described in I. They had been on fairly good pasture until weaned in the middle of October and shipped to Spur. They were kept on a moderate grain and sorghum fodder ration in dry lots for about two months before the initial weights were taken. They were full fed in dry lots for 111 days on a fattening ration of ground milo heads, cottonseed meal, and chopped sorghum fodder.

K. These steers were long-aged calves from the S M S ranch. They were predominantly of Hereford breeding, but several showed very strong traces of Shorthorn blood. They had been driven from near-by pastures and put in the feed lots at Spur on a moderate grain and sorghum fodder ration more than a month before the first weights were taken. Some of them already carried considerable fat. They were full fed in dry lots for 111 days on a fattening ration of ground milo heads, cottonseed meal, and chopped sorghum fodder.

L. These were high-grade or purebred Jersey cows which were being used in feeding experiments. The first experiment is described in Bulletin 352 of the Texas station (*8*). The cottonseed-meal experiments are not yet published. The cows received concentrates in proportion to milk production, a small amount of sorghum silage, an unrestricted quantity of cottonseed hulls, and pasture (either native or oat) whenever it was available. All cows in the experiment

station herd were used in these tests if in suitable stages of lactation and not on register of merit tests.

M. These calves were driven not to exceed one-half mile to the scales. They were on pasture, but were also given hay or corn stover after the first and second day's weighings. They were good to choice Hereford steer calves, obtained in north-central Texas. The cattle and rations are described in detail in United States Department of Agriculture, Department Bulletin No. 1431 (*10*) and in West Virginia Agricultural Experiment Station Bulletin No. 218 (*12*).

N. Driven not to exceed 200 to 300 yards to the scales in the morning after being fed but before being watered. Had been fed wintering rations in lots in an open barn. Same cattle as in M.

O. Same cattle and treatment as in M, except that cattle were a year older.

P. Same cattle and treatment as in N, except that cattle were a year older and lot 3 had received corn silage and wheat straw instead of the grain mixture received by lot 3 the previous winter.

Q. Same cattle and treatment as in M, except that the cattle were now 2 years older and also that after the first day's weighing they were moved a distance of 6 miles. It seems likely that this drive was in large part responsible for the uniformly high experimental errors found at this weighing.

R. Same cattle and treatment as in N, except that the cattle were 2 years older and all had been wintered on the same rations, that is, cottonseed meal, corn silage, and wheat straw.

S. These were steers of all three leading beef breeds. They had been shipped from Mandan to West Fargo and the first day's weights were taken soon after they were unloaded and driven 4 miles to the college cattle yards. They had received neither hay nor water for more than 24 hours before the first weighing. After the first weighing they were grazed on cornfield gleaning and meadow aftermath and were also given some hay.

T. These were steers of all three leading beef breeds. They were on prairie pasture and had been in the pasture where weighed for two days before the first weights were taken and on various near-by pastures for two to three weeks before weighing.

U. These were steers of all three leading beef breeds. They were weighed the first day direct from the stock cars without having had feed or water. They had been shipped from Mandan, where they had been on prairie pasture, to West Fargo and had been in transit more than 24 hours. After the first weighing they were allowed to graze on cornstalk field gleanings and meadow aftermath and were given some hay.

V. These six head were the same as those in U, except that they were not weighed on the first day.

W. Four of these steers were 2-year-old Herefords which had been used in digestion trials; two were 3-year-old Shorthorns in high stocker condition. They were turned out on sweet-clover pasture May 11. Three of them were changed to a light ration of alfalfa hay July 27 and continued on that through September. The four Herefords had been on a light ration before being turned out on the sweet-clover pasture on May 11, and therefore began to make good gains on pasture in about a week or 10 days. The two Shorthorns were turned on pasture directly from a fairly good grain ration and were already carrying a considerable amount of fat. Therefore they lost much weight at first. One of them became adjusted to the change in feed in a few days and regained his loss within the first month. The other continued to lose weight for nearly a week and did not regain all his loss for more than two months. Because of this difference in trends, both steers are omitted from the calculations for May 12–18, as explained in the discussion on pages 572 and 573. These steers are described in more detail in North Dakota Agricultural Experiment Station Bulletin No. 211 (*11*).

X. These steers were of all three leading beef breeds. They had been on cornstalk pasture and had received some hay for about a month just prior to the first day of weighing. They were fed a heavy ration of alfalfa hay and corn silage.

Y. Same as X, except that sweet-clover hay was used instead of alfalfa.

Z. These were steers of all three leading beef breeds. There were four lots, each receiving a different silage in addition to oat straw, cottonseed meal, and corn meal. Silages used were corn, sunflower, millet, and a mixed silage of sweet clover and oat straw.

AA. Same as Z, except that ground barley was fed instead of corn meal and an unmixed sweet-clover silage was fed to one lot instead of the millet silage used the preceding year.

AB. These were steers of all three leading beef breeds. There were four lots, each receiving a different silage in addition to alfalfa, linseed meal, and ground barley. The silages were corn, sunflower, a late cutting of sweet clover, and a mixture of equal parts of an early cutting of sweet clover and an early cutting of corn.

AC. Same as AB, except that corn silage was fed to two lots and sweet-clover silage to two lots.

AD. Same as AB, except that corn silage was fed to one lot, sweet-clover silage to two lots, and salted sweet-clover silage to one lot.

DISCUSSION OF RESULTS

SIZE OF EXPERIMENTAL ERROR

The outstanding conclusion to be drawn from Table 5 is that the experimental error is quite irregular, ranging from as low as 4 pounds to as high as 17 pounds. The average experimental error (regarding each group of animals as equal in importance to every other group, instead of the more laborious but more accurate method of squaring each error, multiplying it by the number of degrees of freedom on which it was based, adding, dividing the sum of these weighted squared errors by the total number of degrees of freedom, and extracting the square root of the quotient) is 10.02 pounds. The frequency distribution of the error is shown in Table 6.

TABLE 6.—*Frequency distribution of different sizes of experimental error*

Size of error (pounds)	Number of groups of cattle	Size of error (pounds)	Number of groups of cattle	Size of error (pounds)	Number of groups of cattle
4.0–4.9	1	9.0– 9.9	11	14.0–14.9	4
5.0–5.9	6	10.0–10.9	8	15.0–15.9	2
6.0–6.9	9	11.0–11.9	11	16.0–16.9	5
7.0–7.9	13	12.0–12.9	7	17.0–17.9	2
8.0–8.9	18	13.0–13.9	6		

MECHANICAL ACCURACY OF SCALES

Care was exercised at all times to see that the scales on which the cattle were weighed were in good mechanical condition. Precautions were taken to see that all bearings and knife edges were free from dirt and that there was no binding of the platform against the frame. A load was weighed in different positions on the scale platform to check the accuracy of the scale. In the 1926–27 experiment at Spur an automatic dial scale of modern design was installed especially for the purpose of assuring the highest possible accuracy in the weights of the cattle, and was tested by using twenty 50-pound standard test weights. Parenthetically, it may not be amiss to observe that while the calves at this weighing showed a very small experimental error, the older group of steers showed an experimental error of 10.5 pounds, which is of about average size.

In only one case was the scale suspected of being not entirely accurate and sensitive to the degree of precision with which the weights were taken. That was the scale on which the dairy cows were weighed. This scale was a wagon scale designed for much larger weights and its knife edges were not in the best condition at

all times. The dairy cows weighed on it showed a larger experimental error than did any other group included in this study, but they were also older and had larger digestive tracts, and it is not at all unlikely that these two latter circumstances were more responsible for the large experimental error than was any mechanical defect in the scale.

Moreover, the method of calculation was such that any error which was constant for an entire day would have been eliminated from the figures. For example, if the weights on any scale had been consistently 10 pounds lighter or heavier than they should have been on any one day, that error would have been eliminated with the daily trend, and hence would not affect the size of the experimental error, which is the subject of this study.

In view of all these facts, it seems safe to conclude that mechanical inaccuracy in the use of the scales had little or no effect upon the size of the experimental errors studied here. However, it might be well to point out that such errors as may possibly have been due in these data to mechanical inaccuracies of weighing will be encountered in other experimental work with cattle unless the workers take even greater precautions than were taken in these experiments to insure accuracy of weights. Therefore the general applicability of the conclusions of this study can hardly be questioned on the grounds of mechanical inaccuracy of the scales, unless it is maintained that the precautions listed above are totally inadequate to insure an accurate weight.

AGE OF CATTLE

In an effort to analyze the causes of the variation in the size of the experimental error, several variables were studied. Age seems to have an important bearing on it, as is shown in Table 7.

TABLE 7.—*Average experimental error for cattle of different ages*

Age of cattle	Number of groups	Unweighted average error (pounds)
1 year and under	12	6.8
2 years and over 1 year	22	9.0
Over 2 years	69	11.0

However, the correlation between age and size of error is by no means close. Thus many of the 2 and 3 year old steers have errors as low as 8 pounds and a few have errors less than 6 pounds. It is safe to conclude, however, that as a general rule the size of the experimental error to be expected increases somewhat with the age of the animals.

WEIGHT OF CATTLE

Weight also seems to have an influence on the size of the error, as shown in Table 8, but this is irregular, and an inspection of the detailed results as given in Table 5 indicates that weight is related to the size of the error only in that average weight and the size of the error are both partially determined by age.

TABLE 8.—*Unweighted average experimental error for cattle of different average weights*

Weight of cattle (pounds)	Number of groups	Average error (pounds)	Weight of cattle (pounds)	Number of groups	Average error (pounds)
300- 399	5	6.4	800- 899	20	12.1
400- 499	5	6.8	900- 999	20	11.5
500- 599	8	9.0	1,000-1,099	16	9.0
600- 699	9	8.5	1,100-1,199	13	10.9
700- 799	4	8.6	1,200-1,299	3	8.6

That it is not weight which influences directly the size of the error is probably shown best by the steers which were fattened in dry lots, as the 13 groups of Kingsville, Big Spring, and Spur steers, which increased their weights by 40 to 115 per cent within four to six months, but showed an average experimental error of 8.8 pounds in their initial weights and an average experimental error of 8.5 pounds in their final weights.

The same thing is shown by the North Dakota silage experiments with fattening steers in dry lots where three-day weights were taken at intervals of 30 days or less. The average errors for the first three-day weights, second three-day weights, etc., are shown in Table 9.

TABLE 9.—*Average error at different periods of fattening, North Dakota silage experiments*

Order of weights	Number of groups averaged	Average error (pounds)	Order of weights	Number of groups averaged	Average error (pounds)
Initial	7	10.9	Fifth	5	9.3
Second	7	9.9	Sixth	4	7.9
Third	7	10.4	Seventh	2	9.2
Fourth	7	10.2			

Here it is clear that there is no tendency for the size of the error to increase with increasing weight. In fact, a straight line fitted to these data shows an average decrease of 0.36 pound in the error from one period to the next. This decrease is on the border line of significance statistically, there being about one chance in thirty that such a decrease could have happened merely as a matter of chance. It seems reasonably certain that there was a real but slight tendency for the error to decrease as the feeding period progressed, in spite of the fact that the average weight of the cattle was increasing rapidly. However, weight was not the only variable which changed with the progress of the feeding period. Doubtless the steers became gentler and less excitable at weighing time as the feeding period progressed, although of that there is no quantitative measure. Possibly a more important variable is the changing nature of the ration, which consisted largely of silage at the beginning, but which was made up more and more largely of concentrates as the feeding progressed. At all events, weight by itself is not a very important factor in determining the size of the experimental error. Such importance as it does seem to show in Table 8 may more logically be attributed to age and to the size of the digestive organs. Quite

possibly there might be found a much closer relation between size of the error and the fat-free live weight instead of the gross live weight, but it seems clear that increasing fatness has no tendency to increase the size of the error.

NATURE OF THE RATION

As mentioned in the preceding section, the increase in the proportion of concentrates in the ration during fattening may possibly be the cause of the slight decrease in the error observed during the progress of the feeding period. It is worthy of note that the errors of many of the groups of animals on pasture were rather large. In contrast to this, however, are such small errors as those of the Mandan steers for May, 1923, and the 1923 fall weights of the West Virginia yearlings which were also grazing on pasture. Moreover, most of the cattle grazing on pasture were older than those in feed lots. The experimental errors given in Table 5 are so irregular with respect to the rations which the different lots received that no clear-cut effect of ration upon the size of the error is evident.

Some of the data have a definite bearing on this point. For example, the Spur 1922–23 2-year-olds were divided into two lots, one of which was fed cottonseed meal and hulls, while the other was fed ground shelled corn, cottonseed meal, and hulls. (The steers and the rations are described in detail in Bulletin No. 309 of the Texas station (*5*).) The rations are distinctly different, especially as to proportion of concentrates included, yet the difference in the size of the experimental error is insignificant, being barely larger than its standard deviation.[6] In the Spur 1926–27 final weights for the Swenson steers there were three lots of steers all receiving milo heads, sorgo fodder, and cottonseed meal. In one lot the sorgo fodder was run through an ensilage cutter and chopped finely before feeding. In another lot the milo heads were fed unground. The lot receiving chopped fodder and ground heads was the most erratic in weight; the lot receiving whole heads and unchopped fodder was least erratic. However, two of the three differences are certainly not significant statistically, and the largest difference is only 2.6 times its standard deviation, and therefore its significance is still open to reasonable doubt.

The final weights of the Big Spring steers also have a bearing on this point when analyzed separately for each lot. (Steers and rations are described in detail in Bulletin No. 363 of the Texas station (*4*) and in Technical Bulletin No. 43 of the United States Department of Agriculture (*2*).) The weights of the steers receiving cottonseed hulls as a roughage were the least erratic in two of the three trials. The weights of the steers receiving sorgo fodder were the most erratic in all three trials. The weights of the steers receiving sorgo silage were least erratic in one trial and intermediate in the other two. However, when the differences in the size of the experimental error are tested for statistical significance, only the difference between the lots fed hulls and those fed sorgo fodder approach significance. That difference is 2.6 times its standard deviation, which makes the

[6] The criterion used in this section for determining significance or nonsignificance of differences in the size of the experimental error is the Z test described in Fisher's Statistical Methods for Research Workers (*3*), p 192–200.

probability that such a difference could have happened by chance slightly less than 0.01.

In the third Kingsville experiment two rations were fed. Each was fed to a lot of Hereford and Shorthorn steers and also to a lot of steers of mixed Shorthorn and Brahman breeding. The rations differed only in the roughage, which was Rhodes-grass hay in one and hegari stover in the other. In the initial weights of the Brahmans, those on the Rhodes-grass hay gave the smaller error (see Table 11); of the non-Brahmans the more erratic weights were obtained from those on Rhodes-grass hay. As the differences barely exceed twice their standard deviation, it is concluded that they are not significant. In the final weights the more erratic are given by the groups getting hegari stover in both cases, the average difference being more than four times its standard deviation, thereby indicating a significant difference. This fact of course does not eliminate the possibility that this real difference may have been due to some other common cause, such as the order in which the lots were weighed rather than to the difference in the nature of the ration. Physically the two rations were not very different, as the hay and the stover were both dry feeds and were chopped into short lengths.

The three lots of West Virginia yearlings in the spring of 1923 show no significant difference in the size of their experimental errors, although their rations during the preceding four months were quite different, two of them including silage and straw, with either clover hay or cottonseed meal, while the other lot received mixed hay and a grain mixture. (A detailed description of the cattle and of the rations used will be found in United States Department of Agriculture Bulletin No. 1431 (*10*).) The West Virginia steers during the winter of 1923–24 all received corn silage and wheat straw. In addition, one lot received clover hay, one received cottonseed meal, and one received mixed hay. Yet the experimental errors found at the end of 120 days in two of the lots on these rations were identical, and the difference between those two and the third barely exceeded its standard deviation.

The weights of the North Dakota steers were not subjected to analysis on this point because the groups on the different rations were small (only five steers in most of them), and the rations in most cases differed only in the kind of silage used, which was usually corn or sweet clover, but also included millet and sunflower silages in some cases.

In surveying this whole group of data on whether or not the nature of the ration affects the size of the experimental error, one is impressed by the fact that nearly all the data show no effect or are contradictory in cases in which the experiment was repeated. Only in the Kingsville data do the figures compel the belief that anything other than chance is needed to account for the observed differences, and even in that case the significance arises almost entirely from the very large error in only one of the lots receiving hegari stover, the last lot weighed; so that it is easy to infer that some special circumstance operated to make the weights of lot 4 significantly more erratic than those of the Brahman lot receiving the same ration or those of the other two lots receiving Rhodes-grass hay. The magnitude of the difference between the fodder-fed lots and the hull-fed lots at Big Spring and

the fact that the difference is in the same direction in all three tests is very suggestive of a real difference. Also the magnitude of the difference between the lot fed chopped fodder and ground heads and the lot fed bundles and whole heads at Spur in 1926–27 is suggestive, although it is based upon only a single test. The writers' conclusion from these data is that the nature of the ration has little influence upon the size of the experimental error, if, indeed, it has any at all.

It should be remembered that all the rations represented in these data were reasonably palatable and not very likely to produce scouring or to cause the cattle to go off feed in any other way. Nothing in these data should be construed as proving that an unpalatable ration, or one which can easily cause scouring and other digestive disturbances in some of the cattle may not cause the weights to be more erratic than weights of similar cattle on a standard ration.

PRECISION OF WEIGHTS

The precision with which the weights were taken seems to have little influence upon the size of the error, as shown in Table 10.

TABLE 10.—*Unweighted average experimental error for cattle weighed with different degrees of precision*

Precision of weights	Number of groups	Average error (pounds)
1 pound	41	9.9
2 pounds	22	10.7
5 pounds	40	9.8

However, the size of the experimental errors found does tell something of the degree of accuracy that is sacrificed by weighing cattle only to the nearest 5 pounds instead of to the nearest 2 pounds or the nearest pound. Fisher (*3*) makes the following statement on this point: "It has been shown that as regards obtaining estimates of the parameters of a normal population, the loss of information caused by grouping is less than 1 per cent, provided the group interval does not exceed one quarter of the standard deviation." Weighing cattle to the nearest 5 pounds is equivalent to weighing them to a much finer degree of precision and then grouping the weights in 5-pound intervals for calculation. What is called "experimental error" in this paper is the standard deviation with which the grouping interval should be compared to determine whether or not the grouping is too coarse. Upon making this comparison it is found that a 1-pound interval is less than one-fourth of the experimental error in every one of the 103 groups of cattle, and therefore that practically nothing would be gained by weighing cattle of these kinds to a still finer degree of precision, e. g., to half pounds. A 2-pound interval is found to be more than one-fourth of the experimental error in 29 of the 103 groups of cattle. Therefore, there is a loss of slightly more than 1 per cent of the information by the use of 2-pound intervals in some of the groups, particularly in the younger cattle kept under the most uniform conditions. However, for most of the cattle a 2-pound interval involves practically no loss of information. On the other hand, the 5-pound

interval is more than one-fourth of the experimental error in all of the groups of cattle, and therefore involves a loss of considerably more than 1 per cent of the information. On the basis of these findings it would seem that cattle should be weighed at least to the nearest 2-pound unit in experimental work, and that if they are young and are being kept under quite uniform conditions, weighing to the nearest pound would be still more accurate. Weighing to units smaller than 1 pound would not result in any additional accuracy in cattle of the ages included in this study, although it might perhaps be advisable in studies of birth weights and other weights of calves up to weaning time.

Weights of groups of steers of course have experimental errors larger, in proportion to the square root of the number of steers in the group, than weights of single steers. Thus a commercial stockyard scale weighing a group of 25 steers in 10-pound units is no less precise than an experimental scale weighing the same steers one at a time in 2-pound units.

In this discussion of precision it has been assumed that the scales used are kept as accurate as their construction will permit and are quite sensitive, at least to the degree of precision used. Now, as a matter of fact, scales are not usually kept in as good condition as that. Dust and dirt get in the bearings, dulling their sensitiveness somewhat; a strong breeze blowing against the scale rack may cause the beam to vary its reading slightly from time to time; the weigher may be in so much of a hurry that he does not always bring the beam to exactly the same position each time before reading the weight. These and other circumstances keep the scales from being exactly accurate, even though research workers generally take every practical precaution to safeguard the accuracy of their scales and of the weights which they obtain. This makes it still more important to weigh experimental cattle with a fine degree of precision. Thus, if the scale breaks slowly, an impatient weigher weighing only to 5-pound units might easily read the weight 5 pounds too high or 5 pounds too low, whereas if he were weighing to 2-pound units he would read it only 2 pounds too high or 2 pounds too low. For this additional reason the writers believe that accuracy is gained by weighing experimental cattle at least to the nearest 2 pounds, and, if they are quite small, to the nearest pound.

The dairy cows used in these experiments were weighed upon a set of old wagon scales which had never been very sensitive, even when new, and the edges had become dulled, so that it was stiff and slow in breaking. This may have been in part responsible for the fact that the weights of these cows showed higher experimental errors than the average of weights taken elsewhere.

BREED AND GENTLENESS OF CATTLE

Complete comparisons between different breeds of cattle are not possible, as usually only one or two breeds were weighed in the same experiment. However, it may not be amiss to point out that the Jersey cows (which were gentle) showed a rather large error, although the size of their digestive organs and perhaps the condition of the scales on which they were weighed probably influenced the size of the error more than any breed characteristics.

The Kingsville weights and all the Spur weights, except the 1922–23 2-year-olds and the 1926–27 Swenson steers, furnish as good evidence as could be desired of the influence (if any) of the gentleness or wildness of the cattle on the size of the experimental error. In all these experiments some of the lots consisted of steers carrying Brahman blood in amounts ranging from three-sixteenths to as much as seven-sixteenths. Other lots weighed on the same days and under the same conditions consisted entirely of high-grade Herefords or Shorthorns. On an average, the Brahman steers were much wilder and more excitable than the other steers, although there was much variation in the disposition of different Brahman steers. Most of the Brahmans improved in gentleness during the fattening period more than the non-Brahmans, but the slight unusual commotion on weighing days was enough to arouse most of the Brahmans to a high pitch of excitement even at the close of the feeding period.

To determine whether the errors actually were greater in the case of the Brahmans, the error was calculated for each lot of steers separately instead of using as one lot all the steers weighed on the same day as was done in the main part of this study. The results of this more detailed treatment of the Kingsville data are given in Table 11.

TABLE 11.—*Experimental error in separate lots, Kingsville data*

Lot numbers in Kingsville weighings	Breed	Experimental error (pounds)	Degrees of freedom
First Kingsville, initial:			
1	Hereford	5.23	72
2	Shorthorn-Brahman	7.36	48
3	Hereford-Brahman	7.25	48
4	Shorthorn	5.50	48
First Kingsville, final:			
1	Hereford	8.39	32
2	Shorthorn-Brahman	4.93	32
3	Hereford-Brahman	5.90	30
4	Shorthorn	7.14	32
Third Kingsville, initial:			
1	Hereford and Shorthorn	5.13	48
2	Shorthorn-Brahman	4.59	48
3	do	5.29	46
4	Hereford and Shorthorn	4.50	48
Third Kingsville, final:			
1	Hereford and Shorthorn	8.31	46
2	Shorthorn-Brahman	6.42	46
3	do	7.93	48
4	Hereford and Shorthorn	14.89	48

When Fisher's Z test of the significance of differences in variance is applied to the figures in Table 11 it is found that in the first Kingsville initial weights the Brahmans were more erratic by a difference which might be expected to occur by chance only about once in four hundred times. On the other hand, in the first Kingsville final weights the Brahmans were the least erratic by a difference which might be expected to occur merely by chance even less frequently than that. In the third Kingsville initial weights the differences are small and probably not significant, although the differences between lots 1 and 3 on the one hand and lots 2 and 4 on the other approach statistical significance. However, those differences do not parallel either breed differences or ration differences, and the writers can not offer a plaus-

ible explanation for them. In the third Kingsville final weights lot 4 is quite significantly more erratic than the other three lots, but the other differences are probably insignificant, the value of Z being less than twice its standard deviation. Thus we see that the Kingsville data are contradictory as to the existence of a breed difference. The fact that the digestive organs of the Herefords and Shorthorns were larger than those of the Brahmans (as revealed by unpublished data obtained at slaughtering time) may account for the observed differences at the time of final weights. The large error for the final weights of lot 4 in the third experiments may be due to the fact that the final weights for this lot were taken at a much earlier hour the third day than on the other two days, so as to permit the cattle to be driven to the loading pens in the afternoon. No very plausible explanation for the higher errors of the Brahmans in the first Kingsville initial weights has been advanced, unless the greater wildness of the Brahmans may be invoked as such.

The Spur data also were recalculated to find the experimental error for each lot separately. The differences in the average experimental errors are shown in Table 12. The cattle called "Herefords" were high-grade Herefords, having probably six or seven top crosses of Hereford blood. Those called "Brahman" were out of the same kind of dams as the "Herefords," but were sired by a three-quarters or by a sixty-one sixty-fourths Brahman bull. They therefore carried a little less than one-half Brahman blood. Those called "back cross" were sired by a Hereford bull out of cows which were half or full sisters to the "Brahman" steers, and therefore the back-cross steers carried a little less than one-fourth of Brahman blood.

TABLE 12.—*Differences in experimental error found for steers of different breeding, Spur data*

Group of weights	Difference in experimental error (pounds)		
	Brahman minus Hereford	Brahman minus back cross	Back cross minus Hereford
1922–23 yearlings, initial	+0.70		
1922–23 yearlings, final	+.02		
1924–25 2-year-olds, initial	−5.03		
1924–25 2-year-olds, final	−5.85		
1924–25 yearlings, initial	+2.30		
1924–25 yearlings, final	+.12		
1925–26 yearlings, initial	−.34	−0.51	+0.17
1925–26 yearlings, final	+1.02	−1.50	+2.52
1925–26 calves, initial	−.99	−3.51	+2.52
1925–26 calves, final	+3.71	+1.19	+2.52
1926–27 calves, initial	−.22	+1.48	−1.70
1926–27 calves, final	+.13	−.61	+.74
Sum	−4.43	−3.46	+6.77
Average	−.37	−.58	+1.13
Probability	Between 0.6 and 0.7	Between 0.4 and 0.5	Between 0.1 and 0.2

The probability expressed in Table 12 is the probability that the observed average difference was not a real one—i. e., that it differed from zero only by chance. This is the test commonly known as "Student's method" of testing the significance of an average difference. From the high probabilities which were found it is obvious

that no one of the three kinds of steers was significantly more or less erratic in weights than either of the other kinds. Detailed examination of the data does show, however, that for some reason (not now apparent) the 2-year-old Herefords fed in 1924–25 were more erratic in their weights, both initial and final, than were the corresponding Brahmans. The probability of the observed difference being due to chance alone is less than 0.05 (Z test) in either initial or final weights alone and much less than that when both are considered together.

The data seem to justify the conclusion that wildness of the cattle or breed differences have little or no consistent effect upon the experimental error of the cattle weights within a lot. It should be pointed out, however, that wildness of the cattle may affect the error of a difference between two lots if one lot contains more nervous cattle than the other. For example, in the initial weights of the 1924–25 yearlings at Spur there was a distinct breed difference in daily trend. The Herefords were gentle, and their second day's weight averaged 5 pounds more than their first, while the third day's weight averaged 12 pounds more than the second. On the other hand, this particular group of Brahmans was the wildest that the writers have handled, and their second day's weight averaged 26 pounds less than their first, and their third day's weight averaged 8 pounds more than their second. Thus the daily trends were different in the two groups, and treating them as one group does not allow for this difference. When they are treated as two groups the differences in daily trend may be subtracted, and the experiental error is found to be 11.4 pounds, with 36 degrees of freedom, instead of 14.9 pounds, with 38 degrees of freedom, as it was when this breed difference in gentleness was not taken into account. This was the most extreme case found of a breed difference in daily trend.

It is believed that the data justify the conclusion that breed makes little difference in the accuracy of the weights, except in so far as it may be associated with differences in the size and capacity of the digestive tract or with differences in nervousness or excitability which are great enough to make the daily trends distinctly different in the lots compared.

UNIFORMITY OF CONDITIONS

Two groups of steers furnish clear-cut evidence as to the effect upon the size of the experimental error of changing environmental conditions. Those are the 64 steers from the Mandan grazing trials in October–November, 1922, and 72 steers from the Mandan grazing trials in October–November, 1923. Both lots were shipped from Mandan to West Fargo, unloaded, driven about 4 miles to the college pens, and weighed directly, having been subject to the excitement and shrinkage of shipment and having been without feed and water for more than 24 hours when the first day's weights were taken. The 64 steers in 1922 were weighed for 6 days, and the experimental error for the whole 6 days (315 degrees of freedom) was 17.7 pounds, as given in Table 5. However, if the first day's weights are discarded, the experimental error for the last 5 days (252 degrees of freedom) is only 12.6 pounds. Likewise, with the 72 head weighed for 8 days in 1923 the experimental error, which is 16.1 pounds when all 8 days are included, falls to 12 pounds when only the last 6 days are included. These facts show clearly that

the shrinkage in shipment did not affect all the steers equally and that a comparison of the weights of different animals, or different groups of animals directly off the cars would have been less accurate than a comparison of later weights.

The six steers in the North Dakota 1921 grazing sweet-clover trials furnish further evidence that animals whose weights are to be compared should be given the same treatment and the same ration for several days before the weights are taken. The four Herefords in this trial were rather thin and had been kept on a light ration for several days prior to May 11, when they were turned out on sweet-clover pasture. At the last moment it was decided that the sweet-clover pasture was so rank that two more steers would be required to keep it grazed down properly. The two Shorthorn steers selected for this use were in fair flesh and had been on a fairly good grain ration until the very day the grazing was started. Consequently, these two lost very much weight just at first, while the four Herefords lost little and began to gain very soon. One of the Shorthorn steers lost heavily at first, but adjusted himself to the new diet in two or three days; the other one lost very heavily at first and did not really begin to gain until about three weeks had elapsed. A measure of the difference in the reaction of the Hereford and the Shorthorn steers to the new ration (sweet-clover pasture) is afforded by the experimental error for the first six days, May 12–18. The error is 12.7 pounds when only the Herefords are included, but is 20.6 pounds when the Shorthorns with their more difficult adjustment to the new ration are included.

The West Virginia steers in the fall of 1924 were driven a distance of 6 miles after the first day's weighing, and the unequal effects of this upon different steers is clearly apparent in the variations in weight and in the three large experimental errors for these steers (13.7, 14.5, and 16.9 pounds). Probably other variations in treatment not recorded in the notes of the experiments will explain a portion of the experimental error in other cases where it is large.

It seems clear from these data that any change in environmental conditions, even though it might seem to apply to all animals equally, is apt to affect some much more than others, and consequently is to be avoided as far as possible in experimental work. It is highly desirable for this as well as other reasons that cattle should be accustomed to their lots, their water, and to the approximate ration upon which they will be fed for at least two or three days before the first weights are taken. It may sound all right for an investigator to excuse a disturbing circumstance in the treatment of his cattle by saying that all animals were subject to the same treatment. But the real question is, Did they all react to it in the same way and equally? The evidence seems clearly to indicate that they usually do not.

Weights of cattle on pasture are not necessarily inaccurate, as witness the low errors of the Mandan steers in May, 1923, and the West Virginia cattle in the fall of 1923, but there is probably more opportunity for disturbing some of the cattle more than others when they must be rounded up from a pasture than when they are merely turned out of a feed lot into the scale pen. Also it is probable that cattle on pasture do not so nearly take all their fill of feed and

water at the same hour of the day as do cattle full fed in dry lots. This may conceivably contribute a little to the inaccuracy of pasture weights for purposes of comparison.

PRACTICAL APPLICATION OF THESE RESULTS TO FEEDING TRIALS

The practical application of the findings of the usual feeding trial is based upon the assumption that the observed differences in gains are caused by the observed differences in feed consumption. That this assumption may not be absolutely true has long been realized. One popular way of avoiding the possibility of error arising from this assumption is to repeat the trial two or more times. Another device not yet very extensively employed is the use of duplicate lots in the same experiment. Other aids are the use of longer feeding periods, larger numbers of animals in the lots, three-day average weights instead of single weights for calculation of gains, refinements in methods of collecting and analyzing waste and refused feeds, etc.

The effect of these different refinements in experimental technic may be better understood if the sources of possible error in the fundamental assumption (that the observed differences in gains are caused by the observed differences in feed consumption) are understood clearly and are measured, as far as possible. Laying aside all questions as to the accuracy of the records of the composition and quantity of the feed consumed and wasted, there is left the question of the accuracy of the observed gains in live weight. Even if the accuracy of the observed gains is granted, there remains the very great question whether the observed differences in gain were caused by observed differences in the ration or were only coincidental with those differences.

This study bears only on the accuracy of the observed weights and of the gains calculated from them. It tells nothing about whether the two groups of steers being compared were in actual fact as evenly matched in their ability to utilize feed as the investigator assumed they were when he finished balancing his lots at the beginning of the experiment. The results of this study showed that the standard deviation of a single weight from its true value was usually from 6 to 12 pounds, being less with younger smaller cattle under absolutely uniform conditions and more with larger older cattle and where there was some disturbance in weather or other environmental condition. Stated in terms of "probable error," this means that in giving a single weight of a single steer under a given set of environmental conditions it should be understood that the actual weight is followed by a probable error of about 4 to 8 pounds.

How large, then, is the probable error of an observed gain in the weight of a steer? If his initial weight and his final weight were each single-day weights, the probable error of the gain would be the square root of the sum of the squares of the two probable errors, or approximately 6 to 11 pounds. If initial and final weights were each the average of three-day weights, these figures would be divided by the square root of 3 and would be approximately 3.3 to 6.5 pounds. Both these calculations are based upon the assumption that environmental conditions would be the same on all weighing days. Since that can not be known in advance and environmental differences can be only partially controlled, it is usually not the absolute gain in

weight of one lot that is studied but the difference between the gains made by the experimental lot and the gains made by the control lot. This removes the necessity for the assumption that environmental conditions are absolutely the same on all weighing days, but it introduces the complication that one is now interested in the probable error of a difference between two gains rather than in the probable error of one gain. Since the errors in weighing are uncorrelated with each other, the probable error of the difference between two gains is equal to the square root of the sum of the squares of the probable errors of the two gains. Thus for two steers each weighed three days at the beginning and three days at the end of the experiment, the probable error of the difference in gain due solely to inaccuracies of weights would be from 4.7 to 9.2 pounds. If there were more than one steer in each lot, as there almost always is, the error due to inaccuracy of weights would be reduced in proportion to the square root of the number of steers in a lot. Thus in the ordinary comparison of the average total gains made by two lots each containing 10 steers, the probable error due solely to inaccuracy in weights would be from 1.5 to 2.9 pounds. Even if there were 25 steers in each lot, these figures would only be reduced to 1 and 1.8 pounds.

With errors of this magnitude in the accuracy of the weights, it should be obvious that differences of less than 5 pounds in the average total gain of two lots of 10 steers are without significance, as they can easily be the result of the intrinsic inaccuracy of cattle weights. It should also be obvious that no accuracy is gained by carrying average weight or average total gain figures into decimal places in publications. Certainly bulletins giving the results of feeding trials would be easier to read if these meaningless decimals were left out of the tables. This is an improvement which could be made in the bulletins of a great many stations. For example, in Texas Bulletin No. 309 (*5, p. 13*), the total gain of lot 3 is given as 386.5 pounds and the total gain of lot 4 is given as 289.1 pounds. Now, the results of the present study (in which the experimental error was found to be 11.5 pounds in the initial weights and 15.2 pounds in the final weights of these particular steers) make it obvious that, so far as the accuracy of the weights are concerned and assuming identical environmental conditions at the initial and the final weighings, the real gain of lot 3 was 386.5 ± 2.2 pounds and the real gain of lot 4 was 289.1 ± 2.5 pounds. With probable errors as large as these, no accuracy would have been sacrificed had the gains been given as 386 and 289 pounds, and very little accuracy would have been sacrificed had the gains for lot 3 been given as "between 380 and 390 pounds" and those for lot 4 as "a little less than 290 pounds." The latter form of statement would have been accurate enough for the popular edition of the bulletin and would certainly have been more easily read and understood by feeders than the table as it was actually presented. The former statement (386 and 289 pounds) would be preferable for circulation among other animal husbandry workers, as it complies with Kelley's rule (*6*) for determining how many figures are significant enough for publication.

Another example of a bulletin containing tables from which meaningless decimals might have been dropped is Texas Bulletin

No. 363 (*4*) which reports the results of the Big Spring tests. In Table 9, for instance, the average total gains for the three lots (basis of feed-lot weights) are given as 378.86 pounds, 310.75 pounds, and 373.55 pounds. Now, the errors actually found in the accuracy of single weights in this test were 5.8 pounds for the initial weights and 8.6 pounds for the final weights, and there were 15 steers in each lot. This means that the average gains, so far as the accuracy of the weights is concerned, really were:

Lot 1, 378.86±1.04 pounds.
Lot 2, 310.75±1.04 pounds.
Lot 3, 373.55±1.04 pounds.

No accuracy whatever would have been sacrificed by rounding off the second decimal place and practically none by rounding off the first decimal place, although the first decimal place is significant according to a strict interpretation of Kelley's rule. The insignificant decimal places were dropped in the summary in Tables 13 and 14, and the bulletin is thereby made simpler and its findings easier to grasp, without at the same time sacrificing accuracy. These examples (which might be multiplied manyfold from the publications of many of our experiment stations) and the findings of this study seem to warrant the statement that in reports of feeding trials with cattle the author who publishes average weights or average total gains carried out into decimal places does not thereby add accuracy to his findings.

In the two examples given above the actual probable errors found were used to calculate the probable error of the accuracy of the total gain. Other investigators can likewise figure the actual error in the weights with which they are working, or they can use the rough rule developed from the findings of this study, namely, that the probable error of the accuracy of a total gain will rarely, if ever, be as low as 1 pound or as high as 3 pounds with lots of steers containing the usual numbers (10 to 15) of animals. The probable error of the accuracy of the average total gain in the usual lot will ordinarily be from 1.5 to 2.5 pounds.

It will be noticed that the writers have constantly referred to the errors discussed in this study as errors in the accuracy of the weight. This is quite different from the probable error of an average weight or an average gain as calculated by the ordinary biometrical formula. The latter is an error of random sampling which assumes that the observed weights are accurate, and is intended to measure the errors which arise from the fact that the steers in any one lot do not all gain the same amount, and therefore that one lot may by "chance" contain a larger proportion of high-gaining steers than another lot does. The application of this ordinary formula to the usual feeding trial is not legitimate, however, because it is based upon the assumption that the steers have been divided into lots at random. Its application is justified only when the man who divides the steers into lots at the beginning of the experiment does so purely at random or when he can see no difference in any of the steers—i. e., when all the steers are alike in gaining ability, so far as the man who divides them can estimate. In actual practice neither of these conditions is fulfilled. The steers are divided so that in the opinion of the man who does the dividing all the lots are equally balanced. A good

steer in lot 1 is balanced by a good one in lot 2, and an inferior one in lot 1 requires that an equally inferior one be selected for lot 2, etc. Such practice in balancing the lots would eliminate all the error of random sampling if the man who balanced the lots had perfect judgment, and if he balanced them with attention only to the gaining ability of the steers, and if he had plenty of steers from which to choose. Not one of these conditions is usually fulfilled completely. The net result is that the actual error of random sampling is less than that given by the ordinary biometrical formula, but is not entirely eliminated. Just how to measure the effect of balancing on reducing the size of the real error of sampling has not, so far as the writers are aware, been worked out. The fact that balancing reduces the error of random sampling was not appreciated when the probable errors in Texas Bulletin 309 (*5, p. 15*) were calculated, nor does it seem to have been appreciated when the extensive study in Illinois Bulletin 165 (*9*) was published. It has, however, often been discussed informally in the last few years by various men interested in applying statistical methods to problems of animal production.

At all events, the error by which to judge the significance of a difference in gains will be less than the square root of the sum of the squares of the error given by the ordinary formula for the error of random sampling and the square of the error of accuracy, but it can not be less than the error of the accuracy of the weights, which is the subject of this study. This study, therefore, establishes the lower limits below which the probable error of a difference in gain can not fall.

Under uniform environmental conditions the error of the accuracy of an average weight varies inversely with the square root of the number of weights on which the average weight is based. Thus if it is impractical to weigh for 3 days, the same accuracy can be secured by using three times as many cattle and weighing only 1 day. Thus the accuracy of the average weight of 30 steers for 1 day should be equal to that of the average weight of 10 steers for 3 days. However, on the one hand, the 30 steers weighed 1 day would have a considerably smaller error of sampling. On the other hand, the 10 steers weighed for 3 days would permit an estimation of the effect of environmental condition and therefore a more accurate estimation of the actual weight of gain of the steers in pounds. This advantage of the 3-day weight is not of importance in comparisons between lots fed at the same time and weighed on the same days, but it would be an advantage in attempts to use the absolute gain in calculating the energy content of the feed or for comparison with gains on similar rations in other years or at other places. Thus a 1-day weight of 30 steers and a 3-day weight of 10 steers are not exactly identical in value for experimental purposes. Each has its advantages, although the probable error of their accuracy for purposes of comparison with similar lots in the same experiment would be identical. If one has a given number of steers and wishes to know what accuracy he will gain by weighing for 3 days instead of 1 day, the findings of this study confirm those published in the preliminary report—that he will reduce the error of the accuracy of his weights for purposes of comparison with other lots in the same experiment to only 58 per cent $\left(\sqrt{\frac{1}{3}}\right)$ as much as it would have been with a 1-day weight, and,

in addition, he will have a still better estimate of the accuracy of the absolute weights and gains for purposes of comparison with weights and gains made in other years or at other places.

CONCLUSIONS

The experimental error (standard deviation) in the accuracy of single weights of cattle may be expected ordinarily under uniform conditions to be between 6 and 12 pounds.

The experimental error is somewhat smaller with younger cattle, and is distinctly smaller under unchanging environmental conditions than where some sudden change is made in some important condition between two of the weighings.

The size of the experimental error is probably not directly related to the gross weight of the cattle, except in so far as both are partially determined by age. The size of the experimental error may perhaps be rather closely related to the fat-free weight of the animal or to the size and capacity of the animal's digestive tract.

Breed seems to have little or no effect upon the size of the experimental error.

Nervousness and excitability of the cattle seem to have little or no effect upon the size of the experimental error when all the steers in a lot are excitable or when all are gentle. Including excitable and gentle animals in the same lot does increase the experimental error.

Weighing cattle to the nearest 5 pounds sacrifices a considerable amount of information. Weighing to the nearest 2 pounds does not lead to any serious loss of information, except with really small cattle. Weighing to the nearest pound adds some accuracy when working with calves and small yearlings. Weighing to units smaller than 1 pound does not seem to possess any advantage which would make the practice worth adopting.

The "probable error" of the accuracy of a single weight under uniform conditions is approximately equal to two-thirds of what has been called in this paper the "experimental error"—i. e., the probable error of a single weight is usually 4 to 8 pounds. The probable error of the accuracy of the average weight of a group may be obtained by dividing the above figures by the square root of the number of steers in the group. Thus, in the ordinary lot of 10 steers weighed for three successive days, the probable error of the accuracy of the average weight would usually be 4 to 8 pounds divided by the square root of 30 (the number of weights), which would be about 1 to 2 pounds. The probable error of the accuracy of a difference between the average weights of two such lots would be about 40 per cent larger, or, roughly, 1 to 3 pounds. With errors of such size no accuracy is gained by carrying into decimal places average weights for the ordinary lot of experimental cattle. It is just as accurate to carry the average weight only to the nearest whole pound, and such a practice would certainly add to the ease with which the published results might be read.

The "probable errors" and "experimental errors" studied in this paper refer only to the accuracy of the weight of the cattle composing those lots, and do not have any bearing on the probable error of the sampling which took place when the animals were selected for those lots. The latter is the probable error given by the ordinary

biometrical formula. Unfortunately, the ordinary formula is not usually applicable to feeding experiments because sampling is not random. The true figure by which to judge the statistical significance of a difference in gains can not be less than the error of the accuracy of the weight (which is the subject of this paper) nor more than the square root of the sum of the squares of the error of accuracy and the error of sampling. The absence of a satisfactory method for estimating the error of sampling which is not random does not at present permit more than a rough estimate of the real error by which to judge the statistical significance of the results of feeding trials. The upper and lower limits between which such an error lies can, however, be rather accurately estimated.

The relative error of a 1-day weight and a 3-day weight is as the square root of one and the square root of one-third when the comparison is being made between the weight changes made by the experimental lot and the weight changes made by the control lot. However, if the experimenter wishes to study absolute changes in weight (as where there is no control lot) or to compare gains made at different times or in different places, the 3-day weight has the additional advantage of permitting the estimation of the day-to-day fluctuation in weight, and hence a more accurate estimation of the absolute weights and gains. For purposes of comparison with the control lot, weighing for 2 additional days eliminates about 42 per cent of the error contained in a 1-day weight. If it is inconvenient or impossible to weigh more than 1 day, 3-day accuracy for comparison with the control lot may be attained by using three times as many animals in each lot. This procedure has the statistical advantage of giving a much smaller error of sampling, but economic reasons usually preclude its use.

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